

ASRC  
UNIT B  
SMALL ANIMAL  
RESEARCH FACILITY

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

UNIVERSITY OF MISSOURI-COLUMBIA  
ANIMAL SCIENCE RESEARCH CENTER  
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Standard Operating Procedure

I. Title: General Information

- A. The ASRC small animal research facility accommodates small, laboratory animals for use by investigators in the College of Agriculture, Food Science and Natural Resources. Other investigators may be assigned space on a cooperative basis for studies of nutrition, genetics, physiology, endocrinology, toxicology and related areas. Space in the facility will be used only for conventional, germ free or low -level pathogen studies.
- B. Operation of the ASRC Small Animal Research Facility is the responsibility of all 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. 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.
- C. 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 for information about standard procedures.
- D. 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.
- E. Individual room assignments will be made on a yearly or short-term basis. In September, users of the facility may apply for space to meet their housing needs for the next year. These applications are reviewed and assignments will be made for continuous (annual) use effective October 1. Short-term use may be requested as needed. Investigators must have a current Animal Use Protocol on file in the ACQA office.
- F. Joint-use rooms are available for all investigators. They must be kept clean and neat for use by the next person.
- G. In order to reduce the risk of spreading disease, it is the facility policy that animals may not be returned to the facility after leaving the building and transported where they have been exposed to other animals or disease causing agents.
- H. Security: Access to the ASRC Small Animal Research Unit will be limited to authorized personnel only. Authorized personnel must accompany all visitors to the Unit.

- I. Housing: Room population density is recommended to be 1 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.
  - 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 dictates otherwise.
  - 4. Albino mice must not be housed on the top shelf of the rack due to the possibility of eye damage from high illumination.
  
- J. Watering: 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 cleanliness. Water bottles are sanitized in the cage washer. Sipper tubes and stoppers are sanitized in the cage washer or soaked in disinfectant solution before being reused.
  - 2. Animals using the automatic water system will be checked daily to assure they are receiving water.
  
- K. Feeding: All animals will have access to food at all times unless experimental procedures dictate otherwise. The food will be clean and free from contaminants.
  - 1. Remove feed that is molded or otherwise soiled.
  - 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 feed milling date, located on the bottom of the feed bag, must be recorded on the lid of the feed barrel when the new feed is placed in the barrel. Mouse, rat, hamster and rabbit feed should be used within 90 days of opening. Guinea pig food should not be used if older than 90 days without vitamin C supplementation.
  - 3. Feed should be used in a manner to prevent excessive storage. Unopened bags with a milling date of over 6 months need to be discarded.

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II. Title: 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 facilities 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 care of animals;
6. Recommending suspension of activities involving animals that are not in compliance with standards; and
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 all 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, and the College of Veterinary Medicine. 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 has required that institutions, including MU, receiving PHS funding assure the humane care and use of animals. This compliance takes the form of an Animal Welfare Assurance statement filed with, and approved by, the Office for Protection from Research Risks. MU's assurance includes a statement of policy to comply with appropriate laws and guidelines; a listing of facilities; detailed 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.

Standard Operating Procedure

III. Title: Personnel

- A. Treatment and handling of animals will be conducted in a humane manner.
- B. It is the responsibility of the investigator to provide adequate personnel to properly maintain his/her animals during both the week and weekend hours including holidays. Proper maintenance includes feeding, watering, cage changing, cage washing and room sanitation.
- C. Orientation programs will be scheduled throughout the year as needed. Upon assignment to the Small Animal Research Facility, new students and employees should report to the Research Animal Facility Manager to make arrangements for attending an orientation session.
  - 1. Individuals working with animals or equipment within the Small Animal Research Facility are required to complete the orientation program prior to 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, drinking, and smoking are not permitted in animal or service rooms. A break room is provided in the facility for eating and drinking.
  - 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.
- D. Any employee bitten by an animal or injured on the job should report immediately to their supervisor who will arrange for medical attention and completion of accident reports.
- E. Persons conducting activities in the ASRC Small Animal Facility are required to participate in the MU Occupational Health Program. It is the responsibility of the Investigator to arrange for their staff to be included in this program. Information is available on the Environmental Health and Safety website.

Standard Operating Procedures

IV. Title: Procurement

A. Animal Procurement

1. The Facility Manager places all orders for animals to be housed in the ASRC Unit B Small Animal Facility. Forms to request animals are available in the Unit office and on Group Share.
2. Investigators wishing to initiate a protocol using animals are required to fill out a new Animal Care and Use Protocol Review Form. These forms can be obtained by downloading them from the Office of Research website:  
[www.research.missouri.edu/research/forms/local\\_index.html](http://www.research.missouri.edu/research/forms/local_index.html).
3. An original protocol must be on file in the ACQA office. This protocol must include special information pertinent to the management, health care and use of the research animals. This form establishes the records required by regulatory and granting agencies. In addition, it establishes a permanent protocol number, which will remain assigned to that project until its completion. Protocols using carcinogens, toxins, radioactive materials or biohazardous material must establish a project safety protocol with the Health Physics Department.
7. A room request form must be completed and approved by the Unit Supervisor before animals are placed in the facility. In addition, the Attending Veterinarian and Facility Manager must approve the health of the animals before animals are housed in the facility. Animals with pinworms, fur mites, Mouse Hepatitis Virus and other contagious diseases will not be allowed into the facility.
8. All animals used in the ASRC animal facility must be from approved sources.  
(See VIII.) Animals will be procured only when adequate cages and space are available.

The following are sources of laboratory animals routinely used by the ASRC. Other sources are used at the 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. The investigator will be responsible for all costs incurred while the animals are housed outside of the ASRC.

### 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.  
P.O. Box 29176  
Indianapolis,IN 46229  
(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.  
P.O. Box 29176  
Indianapolis,IN 46229  
(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

## B. Supply Procurement

1. Feed and bedding will be ordered and purchased by individual 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). 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, 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 long-term use.
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 other facilities on campus.
3. Conversion to usage of plastic cages for mice and rats necessitated implementation of a fee structure to pay for replacements. The fund, initiated in July 1998, helps pay for cage replacement costs and costs for future caging needs. Investigators will be charged on a per diem basis for all shoebox cages being used in their room.

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V. Title: Animal Health Monitoring

- A. The most important aspect of animal health monitoring is frequent observation of all animals. The condition and welfare of all animals should be monitored daily and noted on the Animal Health Observation sheet in the animal room. 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. These must be recorded on the "Door Sheet" of the animal room.
1. Any animal found out of the cage is to be captured and placed in a clean, separate cage. The principal investigator is to be notified.
  2. Any animal showing any abnormality, i.e., grossly sick, injured, lame, circling, head tilt, loss of appetite, etc. must be noted on the Animal Health Observation sheet and initiate an animal health card (blue card) if necessary.
    - a. In the event a health problem is observed during the monitoring, the Principal Investigator or the Facility Manager is to be notified.
    - b. The OAR Veterinarian on veterinary care rotation will be called if treatment of the animal is necessary.
    - c. Contact Procedure: During normal business hours (8 am-5 pm) use pager number 441-4141. During evenings, weekends, and holidays use pager number 441-4198.
    - d. Report abnormalities even though they may seem to be of minimal significance, and regardless of whether experimentally-induced or spontaneous.
  3. Any animal found dead will be recorded on the Animal Health Observation sheet. Dead animals are to be taken to the freezer in room 126 or walk-in freezer, N192.
  4. Animals should be observed daily, before 1:00 p.m., with the Animal Health Observation sheet filled out and signed. A Laboratory Animal Clinical Record (Blue Card) should be initiated for each sick animal. Cards are available outside the Facility Manager's office (N154). The card should be clipped on the door with the Animal Health Observation sheet.
- B. The following information should be filled out on the blue card:
1. Species or Strain
  2. Animal/cage number of the ill or injured animal.
  3. Number of animals observed to be ill or injured.
  4. Sex of the animal
  5. Age of the animal - if the age is unknown, state whether the animal is a suckling, juvenile or adult.
  6. Protocol number to which the animal is assigned.
  7. Room #
  8. Investigator Name and Phone #
  9. Date and observed abnormality

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VI. Title: 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 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 animals (alive or dead), tissues, samples, or swabs, must be accompanied by a RADIL submission form; forms may be obtained from the Facility Manager. All submissions 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. Indicate type of profile requested (Clinical, Basic, or Comprehensive profile).

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

1. The OAR will supply animals for 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 120 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.

3. Each time the bedding is changed, soiled bedding from other cages in the room is mixed with clean bedding for the sentinel cage. Individuals changing cages should be sure to sample dirty bedding from each shelf on a rotating basis.

#### E. Rodent Vendor Surveillance

All rodents coming into the facility must be from an approved vendor or source specifically approved by the Attending Veterinarian. Vendor approval is based on companies approved by the ASRC. Acceptance of animals into the facility will be based on review of health status by the Attending Veterinarian, Unit Supervisor and Facility Manager. All rodents coming into the facility from a source other than a commercial vendor will be quarantined at the OAR Medical Center facility before they can be housed at the ASRC Unit B facility.

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VII. Title: 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 will display a cage card containing the following information: species, number of animals in cage, investigator's name and phone number, source from which animals were obtained, date of arrival or birth and USDA number where applicable.
  
- 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.
  
- F. Radioactive Materials and Biohazards: Use of radioactive materials in The ASRC Small Animal Research Facility must be under the advisement of Environmental Health and Safety.
  - 1. Animal care personnel must be familiar with proper handling procedure.
  - 2. Rooms in which these materials are used must be properly identified.

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VIII. Title: Procedures for Incoming Animals

- A. The Facility Manager will place all orders for animals needed in research projects at the ASRC. A commercial supplier with known clean health status must supply any animals entering Unit B. Animals from other sources must have a health status report on file with the Facility Manager for veterinarian review and approval BEFORE enter Unit B.
- B. Health monitoring reports for all rodents from non-approved vendors are reviewed by the attending veterinarian and /or diagnostic samples are submitted. These animals are quarantined in OLAM facilities in the School of Medicine or College of Veterinary Medicine until satisfactory animal health reports are obtained.
- C. Rodent shipping boxes must be checked by the animal care personnel for damage and dead or sick animals. Dead and sick animals must be removed from shipping boxes and the facility manager notified as soon as possible so that the responsible individuals may be notified.

Ordinarily, rodents should be housed upon arrival, as soon as possible. If this is not possible, they should be placed in their shipping containers, in the room to which they are assigned. Water must be provided by placing a clean water bottle on the top and pushing the sipper tube through the screen.

- D. Quarantine and Conditioning: It is recommended animals should be allowed a minimum period of two weeks for conditioning and health status evaluation before assignment to experiments. Incoming animals suspected of being sick are to be removed from the group and isolated from other animals. The Facility Manager is to be notified of this condition and a blue card will be initiated and the attending veterinarian contacted.

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- IX. Title: Cage Changing Procedure for Contact Bedding
- A. Bedding in direct contact with animals is to be changed once a week. Heavily soiled cages will be changed more often if necessary. DO NOT house animals on top shelf of cage rack.
  - 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-bar lid should be changed every two weeks.
    - 7. Micro-isolator tops, if used, should be changed when it becomes visibly dirty.
    - 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 and 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 hallway.
  - D. Dump bedding in a bag-lined dumpster. Any food, bedding, or excreta remaining in the cage must be thoroughly scraped free. Seal liner and remove to the dumpster located outside the dock.
  - E. Pre-wash all cages and equipment. Remove all tape and cage cards.
  - F. Stack equipment neatly in dirty cage wash area.

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- X. Title: 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.
    - a. Poultry pans typically use brown pan paper, which is located in room N164. It is purchased by the investigator.
    - b. Tear paper off of roll using the pan in front as a size guide. Too much paper will cause the paper to crumple when inserted into 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 once every two weeks.
  7. Food hoppers should be changed once 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. Remove all tape and cage cards.
  3. Load pans on to rack used to wash pans in cage washer or stack neatly in cage washing area.

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XI. Title: Room Maintenance

A. Floor cleaning schedule

1. All floors in animal rooms are to be swept daily with brooms provided.
2. All floors in animal rooms are mopped with a disinfectant solution when cages are changed. Make sure the proper dilution is used. Pour mop water down drain once a week to keep drain trap from becoming dry.
3. Mop buckets should remain outside the room while mopping.
4. All floor cleaning equipment is to be stored in a clean condition in the cage washroom.

B. Sink cleaning schedule

1. Sink and counter area are to be cleaned bi-weekly with detergent 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 Manager will periodically designate a time for this during the year.

D. Any environmental problems in any rooms, i.e., temperature, humidity, leaking faucet, etc., will be reported to ASRC personnel.

1. Timers are for each animal room to provide automatic light control. Users are to contact the Facility Manager for setting and operating the timers.
2. Light bulbs will be replaced by maintenance after being reported to the Manager.

E. Air filters should be changed once a month or more often if necessary.

Standard Operating Procedure

XII. Title: Cage 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.
3. There will be a charge for any equipment left in dirty cage wash area for over 48 hours without being pre-washed. (Cages and pans-5 dollars each, racks-25 dollars each)
4. 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 goggles, gloves, lab coat, and boots must be worn during acid-cleaning activities.
2. Lab coats and shoe covers are available in the break room (N156).

C. Sanitization of Equipment - **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 day after cage washer has reached recommended temperature (180°F).
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.
4. 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.

Standard Operating Procedure

XIII. Title: Summary of Animal Care and Cleaning Schedules

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 with finger 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 Procedure

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.

Standard Operating Procedure

XV. Animal Space Requirements and 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: 5 mice weighing <25 grams  
4 mice weighing  $\geq 25$  grams
- Large cage: 12 mice weighing < 25 grams  
9 mice weighing  $\geq 25$  grams

## B. Handling

1. Mice may be easily transferred from cage to cage by gently lifting them by the base of their tails.
2. Mice can be restrained by gently but firmly pinching the skin on the back of their necks and wrapping the tail around a finger.

## C. 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.
3. 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.

## C. 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.

## E. 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.

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

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.

### B. 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.

### C. 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.

#### D. 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.

#### E. 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.
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.  
(Temperature 70-80°F and Humidity 45-55%)

### C. 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.

### D. 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
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.

### E. 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.

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

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

### C. 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.

#### D. 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.

#### E. 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 stand erect
Quail ----	.25 ft <sup>2</sup>	
<0.25 kg	.25 ft <sup>2</sup>	with feet on wire
Chickens <0.5 kg	.50 ft <sup>2</sup>	
0.5-1.5 kg	1 ft <sup>2</sup>	
& 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<sup>1/2</sup> X 9<sup>1/2</sup> X 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

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.

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

### B. 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.

### C. Health Problems

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.

### D. 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 Procedure

XVI. 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.

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

Individual 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.

## Standard Operating Procedure

### XV. 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. Group or singly housed animals should 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.

- 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.

#### **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 Rodent Sentinel Program**

**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 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 animals.

**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 pick-up previously unknown pathogens, monitor known pathogens and to evaluate the overall health status of the rodent colonies on campus as a whole.

### **Procedures:**

#### **1. Placement:**

- Before the sentinel placement, two mice and two rats from each new sentinel group will be submitted to a diagnostic laboratory for comprehensive necropsies, including fecal PCR for *Helicobacter* sp. The remaining animals are placed in a barrier room pending the results of the necropsies.
- Sentinels will be supplied to each animal room, and housed in a shoebox cage(s) on the bottom shelf of one or more of the racks. The standard stocking rate for sentinels will be one cage of two sentinel animals for approximately every 120 cages in each animal room. The Assistant Director of OAR, must approve exceptions to stocking rate.
- In most cases, 6-8 week old females will be supplied quarterly to each room. Male sentinels will be placed instead of females if specifically requested by the investigator.
- Mice used, as sentinels are CD-1 albinos ordered from Charles River. Rats used as sentinels are Sprague-Dawley from Charles River.
- 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 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.

#### **2. Pre-placement Submission:**

- If the sentinel animals are declared free from infectious disease, they are placed into animal rooms as sentinels. Normally sentinels remain in the animal rooms for 8-10 weeks before being submitted to a diagnostic laboratory.

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

- At each bedding change, a dirty cage in the room will be identified as the new sentinel cage.
- Into this dirty cage, a single scoop of dirty bedding (from a one (1) oz. disposable plastic cup) will be added from each cage on the shelf(s) scheduled for sampling that week. After all scheduled cages have been sampled; the volume of bedding will be adjusted by adding additional dirty bedding, removing excess dirty bedding, or by adding no more than two (2) scoops of clean bedding.
- Each sentinel cage will have a sentinel cage card that will be used to keep track of which shelves have been sampled.

#### 4. Submission Schedule

The schedule for submissions from each room classification (see **Room Classification SOP**) is provided below. Sentinel monitoring is performed on a quarterly basis for all animal rooms.

ROOM CLASSIFICATION	QUARTER			
	1	2	3	4
A	CS + fecal	Comp. Necropsy	CS + fecal	Basic sero + fecal
B	CS + fecal	Comp. Necropsy	CS + fecal	Basic sero + fecal
C	CS + fecal	Comp. Necropsy	CS + fecal	Basic sero + fecal
D	CS + fecal	CS + fecal	CS + fecal	CS + fecal
E	CS + fecal	CS + fecal	CS + fecal	CS + fecal
F	CS + fecal	CS + fecal	CS + fecal	CS + fecal

\* See below note regarding *Helicobacter free* animals.

- **CS = RADIL Clinical Serology** panel.
  1. In **mice**, this includes testing for the presence of antibody to MHV, MMV, MPV, Sendai virus, TMEV, EDIM, and *Mycoplasma pulmonis*.
  2. In **rats**, this includes testing for the presence of antibody to RCV, Sendai virus, PVM, Parvo, TMEV, and *Mycoplasma pulmonis*.
  
- **Fecal** = Fecal flotation and cellophane tape test, as well as examination of the pelage for evidence of external parasites.
  1. Samples collected and evaluated by the CLAT at MSB, (Sherrie Neff).
  
- **Comp. Necropsy** = RADIL **Comprehensive Necropsy** panel.
  1. In **mice**, all sentinel mice are tested for ecto and endo parasites, their respiratory tract is cultured for the presence of *Pasteurella pneumotropica* and *Mycoplasma pulmonis*. Their GI tract is cultured for the presence of *Salmonella spp.*, and serology is performed to detect the presence of antibodies to MHV, MMV, MPV, Sendai virus, *Mycoplasma pulmonis*, TMEV (GDVII), EDIM, PVM, REO3, LCM, Ectromelia, MAD1, MAD2 and Polyoma.
  2. In **rats**, all sentinel rats are tested for ecto and endo parasites, their respiratory tract is cultured for the presence of *Pasteurella pneumotropica* and *Mycoplasma pulmonis*. Their GI tract is cultured for the presence of *Salmonella spp.*, and serology is performed to detect the presence of antibodies to RCV, Sendai virus, PVM, Parvo, *Mycoplasma pulmonis*, TMEV (GDVII), REO3, LCM, MAD1, CARB, Hantaan and Tyzzer's.
  
- **Basic sero** = RADIL **Basic Serology** panel.
  1. In **mice**, this includes testing for the presence of antibody to MHV, MMV, MPV, Sendai virus, TMEV, EDIM, PVM, Reo3, LCM, Ectromelia and *Mycoplasma pulmonis*.
  2. In **rats**, this includes testing for the presence of antibody to RCV, Sendai virus, PVM, Parvo, TMEV, Reo3, LCM, and *Mycoplasma pulmonis*
  
- **\* Helicobacter Free:** Sentinel animals from colonies that are **Helicobacter free** will be tested by fecal PCR for the presence of *Helicobacter spp.* Exception, are rooms classified as *Helicobacter* positive will not have those sentinels evaluated for *Helicobacter*.