Contents
Welcome ....................................................................................................................................................... 4
Contact Information .................................................................................................................................. 5
Facility Access ............................................................................................................................................... 6
Entering & Exiting the Vivarium ................................................................................................................ 6
Pathogen Free Animals and Disease Status .......................................................................................... 6
Order of Entry ....................................................................................................................................... 6
Dress Code ................................................................................................................................................ 7
Working with Immune Compromised Animals ..................................................................................... 7
Working in Conventional Rooms .......................................................................................................... 7
Entering Chemical Hazard and Biohazard Rooms ............................................................................... 7
Exiting the Vivarium .............................................................................................................................. 7
Transporting Animals ............................................................................................................................ 8
Escaped Animals ................................................................................................................................... 8
Animal Environment (Guide pp. 41-104) ...................................................................................................... 8
Macroenvironment (Animal Room) ........................................................................................................... 8
Procedures ............................................................................................................................................... 8
Illumination ........................................................................................................................................... 9
Temperature and Humidity .................................................................................................................. 9
Ventilation ............................................................................................................................................. 9
Sanitation .............................................................................................................................................. 9
Pest Control ........................................................................................................................................... 9
Storage .................................................................................................................................................. 9
Waste Disposal .................................................................................................................................... 10
Microenvironment (Animal Cage) ........................................................................................................... 10
Animal Identification ........................................................................................................................... 10
Housing Density .................................................................................................................................... 10
Environmental Enrichment .................................................................................................................. 10
Sanitation .............................................................................................................................................. 11
Bedding (Guide p. 68)............................................................................................................................ 11
Feed (Guide p. 65) ................................................................................................................................ 11
Water (Guide p. 67) ................................................................................................................................ 11
Animal Monitoring ................................................................................................................................. 11
Welcome

This handbook has been prepared to provide information and guidelines for anyone currently using or planning to use animals in the SIU vivarium. This may not cover all your questions, so please feel free to ask the people listed in the section below.

The Institutional Animal Care & Use Committee (IACUC) and the Attending Veterinarian (AV) have oversight over all animal care and use at Southern Illinois University in accordance with the Animal Welfare Act, the Guide for the Care and Use of Laboratory Animals (the Guide), and the Guide for the Care and Use of Agricultural Animals in Research and Teaching (the Ag Guide). Animal housing and research areas must be inspected and approved by the IACUC prior to use and are subject to routine semi-annual and random inspections while in use.

Standard animal caging, feed, lighting, temperature, and humidity are provided by the vivarium. A research project may require the Principal Investigator manage some of the animal care (for example, when an animal is housed in a laboratory space). These arrangements should be discussed with the Vivarium Director and Animal Care Coordinator and approved by the IACUC prior to receiving animals and starting the research project. Once responsibilities have been established, the project’s animal care plan should be described in a written husbandry Standard Operating Procedure (SOP). The husbandry SOP can then be used for reference and for training new research staff. Any husbandry exceptions to the Guide or Ag Guide must be included in the IACUC protocol application as well as the SOP and approved by the IACUC.

The SIU Laboratory Animal Program (LAP) office is staffed Monday through Friday from 8 am – 4 pm. Caretakers are present daily, 365 days per year. The Vivarium Director or backup veterinarian is available by phone for emergencies after hours, on weekends, and holidays. The vivarium staff is available by appointment to assist researchers with special needs.

The institutional Animal Care & Use Committee membership includes a veterinarian, scientific and non-scientific SIU faculty and staff, and at least one community member not associated with SIU. The IACUC coordinator is located in the Office of Research Compliance in 350 Woody Hall and by phone at (618) 453-4533 or at iacuc@siu.edu. Meeting times and other IACUC resources are available on the IACUC website: https://orc.siu.edu/animal-research/index.php.

This handbook describes vivarium policies and investigator responsibilities when housing laboratory animals at SIU. More detailed information on animal care can be found in the Guide for the Care and Use of Laboratory Animals (the Guide).
Contact Information

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Facility Access

The vivarium is kept secured and access is restricted to authorized personnel only. Only animal care staff or research personnel who are listed on approved protocols are granted access to the vivarium. Those personnel must have completed the IACUC-required appropriate training and have enrolled in the occupational health program for SIU, in addition to completing an orientation to the vivarium.

Children, pets, and service animals are not permitted in animal facilities. Any persons not listed on an approved protocol must be accompanied by a research staff or vivarium staff member at ALL times and be approved by the Vivarium Director or the Animal Care Coordinator as a visitor.

DO NOT open facility doors to persons unknown to you or prop doors open to allow access. Report any suspicious persons to vivarium personnel. Report lost or stolen keys or fobs to vivarium administration or the LAP office immediately.

Upon IACUC approval of new personnel, contact the LAP office to schedule orientation. Facility orientation will be scheduled for new investigators and their approved research staff as needed.

Entering & Exiting the Vivarium

Pathogen Free Animals and Disease Status
The majority of rodents housed in the vivarium are bred and raised under strict hygienic conditions and are free of pathogens that can impact research variables or that can be transferred to people. Rodents raised as such are called “specific pathogen free” or SPF rodents. To prevent disease outbreaks and significant research disruption, investigators must contact vivarium management for approval before moving animals between rooms and institutions. Animals that are brought into the facility without prior approval may be relocated or euthanized.

Order of Entry
The vivarium has a specific order of entry to rooms that must be followed to maintain biosecurity. The order of entry is posted in the vivarium beside Room 1073. The cage change schedule is also provided. Always check the posted order of entry as it can change frequently. In general, known-infected and quarantine rooms are entered last. As a best practice, please do not enter any area not directly related to your work.

Care staff make every effort to keep extra supplies stocked in each animal room, but not every need may be anticipated. If you have need for supplies that are not stocked in your animal room, please ask care staff for these items. The storage areas are considered restricted access and must only be entered by care staff.
**Dress Code**

To enter the vivarium, you must wear shoes with covered toes and have legs covered to the ankle. Labcoats are required except for care staff that are wearing dedicated clothing and shoes. Strict labcoat hygiene should be observed in keeping with good laboratory practice. Visitors will be supplied with labcoats and shoe covers if necessary.

**Working with Immune Compromised Animals**

Before entering the room, don a disposable gown, exam gloves, and a surgical mask. Only one researcher may work at the transfer station at a time. Lightly spray the transfer station with the disinfectant provided and wipe down with paper towels, keeping contact times in mind. Open all cages in the transfer station and lightly spray your gloved hands between every cage. When work is complete, lightly spray the transfer station with disinfectant and wipe down with paper towels. Immediately before exiting the room, remove your gown and gloves and wash your hands.

**Working in Conventional Rooms**

In addition to the clean lab coat, wear exam gloves and a surgical mask. Only one researcher may work in the room at a time and use should be coordinated in advance to reduce conflicting needs. Lightly spray the work surface with disinfectant and wipe down with paper towels, keeping contact time in mind. When work is complete, lightly spray the work surface with disinfectant and wipe down with paper towels. Remove gloves and wash hands prior to exiting the room.

**Entering Chemical Hazard and Biohazard Rooms**

Animal projects with ABSL-2 hazards require approval through the Institutional Biosafety Committee. Vivarium staff must be notified at least one week before the hazard project is to begin. Complete a hazardous projects form and submit it to the LAP office by email. Rooms with hazardous materials must be properly labeled and the project specific SOP must be posted on the door. Anyone entering the room must follow the safety procedures within the SOP including personal protective equipment, animal containment, and waste handling. PPE and handling requirements will vary between projects. Please reference the appropriate LAP SOP for each condition.

**Exiting the Vivarium**

PPE (gowns, gloves) should be removed prior to exiting individual rooms (unless also utilizing the necropsy room prior to leaving the vivarium). Hands should also be washed prior to exit.

Do not wear gloves in public corridors. If gloves are necessary to transport materials, wear one glove to handle the transported item. Your free, ungloved hand is then used to touch doors, elevator buttons, etc. Gloves worn in corridors give others the impression of contamination.
Transporting Animals
Investigators must have approval from the IACUC to move animals outside of the vivarium to IACUC approved spaces. Proper procedures to transport animals include:

When transporting rodents through public corridors or between facilities, use a cart when handling more than one cage per person and avoid stacking. Shipping containers designed for stacking may be stacked at a maximum of two cages. There must be a means to prevent unintended opening such as a latch or locking mechanism, tape, rubber band or a box-within-a-box. Use covered or opaque cages/carriers or use a clean, breathable drape; alternatively, cages could be placed in a secondary opaque, but vented container (e.g., plastic tub with air holes). Rodents should be transported within filter-topped enclosures to reduce the transfer of allergens. Never leave animals unattended outside of an animal housing or use area.

Escaped Animals
Any animal that gains access to the floor or walls or otherwise escapes enclosure must be reported to the Animal Care Coordinator immediately. Once captured, the animal should be placed into a separate cage and placed separately on the rack. Alternatively, the animal may be euthanized immediately.

Animal Environment (Guide pp. 41-104)

The Guide provides detailed recommendations for the lighting, temperature, humidity, cage sizes, cage density, feed, water, bedding materials, sanitation frequency, outdoor housing, and behavioral management for agricultural, terrestrial, and aquatic animals. Exceptions to the Guide recommendations must be described in the IACUC protocol application and approved by the IACUC. Examples of exceptions include significant changes in circadian cycles, limited feeding, and single housing of social animals.

Macroenvironment (Animal Room)

Procedures
When working with or around animals, work quietly to avoid disturbing or distressing them. Any stressful procedures, such as surgery or euthanasia must be done outside of the animal housing rooms to avoid unnecessary stimulation of the other animals.
**Illumination**
Light cycles within the animal rooms are controlled by timers. The standard light/dark cycle is 12:12 in rodent rooms. Significant variation requires project-specific IACUC approval. Red lights are available for dark-cycle use in some rooms.

**Temperature and Humidity**
The temperature and humidity in indoor housing rooms must be monitored and recorded daily. The thermostat is set at 70°F. Animal housing rooms should be able to maintain room temperature within +/-3° of the room’s set temperature. The relative humidity levels for indoor animal rooms should be at least 50% +/-20%. Levels will sometimes drop below 30% in the winter. Measures will be taken to increase the humidity if there are any clinical effects or if research may be impacted. The recommended temperature range for rodents is 68-79° (Guide p. 44).

**Ventilation**
The broad guideline is 10-15 air changes per hour, but more or fewer may be necessary depending on room density and characteristics of the air supply (Guide p. 46). Assessment of air changes is conducted yearly or more often if performance standards are in question.

**Sanitation**
Animal rooms and support areas must be cleaned and disinfected regularly to minimize contamination and harborage of vermin (Guide p. 72). At minimum, floors and countertops are cleaned and disinfected at time of cage cleaning. More frequent cleaning may be performed when deemed necessary. Cleaning activities are documented on a room log with date and initials of personnel performing the task. Husbandry staff will clean the room but will not clean investigator equipment within the room – these items are the responsibility of each investigator and may only be kept within the room with prior approval from the Animal Care Coordinator. Sanitation efficacy is assessed quarterly by the LAP sanitation monitoring program.

**Pest Control**
The animal housing areas must be kept neat and clean to prevent the harborage of vermin. Any feed, nutritional enrichment, or treats must be stored in vermin-proof containers and labeled with an expiration date. Any evidence of insect or wild rodent infestation should be reported to LAP staff as soon as possible.

**Storage**
The storage of unnecessary supplies, equipment, and materials should not occur in animal housing areas. Clutter makes the area difficult to sanitize and can attract vermin. Do not use cardboard boxes in animal areas because they cannot be sanitized and may harbor insects. Exceptions require approval of the Animal Care Coordinator.
Waste Disposal
Vivarium staff and Investigators are responsible for proper disposal of trash, animal waste, carcasses and any hazardous waste within the animal areas. If you have any questions about proper disposal of items, please contact vivarium staff or the Center for Environmental Health and Safety (CEHS).

Microenvironment (Animal Cage)

Animal Identification
Animals should be clearly identified through the use of cage cards that include the name and contact information of the investigator, protocol number, source, date received/weaned, birthdate, gender, number in cage, and genotype/strain.

Housing Density
The Guide provides detailed recommendations for cage sizes and housing density for single-housed, group-housed, and breeding animals (Guide p. 57). Exceptions to the Guide recommendations required for a research project must be described in the IACUC protocol application and approved by the IACUC. Examples of exceptions include single housing of social animals and having more than one mouse litter in a standard size mouse cage.

- Standard mouse cage size: 77.66 sq. inches
  - Maximum mouse cage density: 5 adult mice or 2 adults (male and female, or two nongravid females) + 1 litter
- Standard rat cage: 140.12 sq. inches
  - Maximum rat cage density: 2 adults or 1 female + 1 litter

Environmental Enrichment
SIU IACUC Policy 500 Environmental Enrichment details the basic behavioral management expectation for animals. Exemptions to environmental enrichment and social housing of social species requires justification and approval by the IACUC. The policy is available online. The vivarium standard for enrichment for rodents is:

- Socially housed mice: cotton nestlet +/- igloo
- Singly housed mice: cotton nestlet +/- igloo
- Socially housed rat: crinkle paper nesting material
Singly housed rat: crinkle paper nesting material

Sanitation

Individually Ventilated Cages are changed and sanitized weekly. Conventional cages are changed and sanitized once or twice per week, dependent upon cage density. Water bottles and food are checked daily and changed weekly for all types of housing (Guide pp. 70-71). Cleaning activities are documented on a room log with the date and initial of the personnel performing the task. Maximum temperature of the cage washer is evaluated weekly. Sanitation effectiveness is evaluated quarterly through the sanitation monitoring program.

Bedding (Guide p. 68)

Both aspen chip and corncob bedding are utilized in the vivarium, based on housing type. Corncob bedding is reserved for use in Individually Ventilated Cages, while aspen chip bedding is used in conventional cages.

Feed (Guide p. 65)

Purina LabDiet 5008 is used for both mice and rats that do not require autoclaved feed. Animals requiring autoclaved feed receive Purina LabDiet 5010.

Water (Guide p. 67)

Animals have constant access to potable, uncontaminated water. Water bottles are checked daily and are changed and sanitized weekly or more often. Research protocols that limit access to water must have prior IACUC approval.

Animal Monitoring

Vivarium management will ensure that trained personnel monitor the animals every day, including weekends and holidays. If a research project requires the research staff to monitor the animals themselves, the PI must review the distribution of responsibilities with the Animal Care Coordinator and record the agreed-upon duties on the Investigator Husbandry Log.
Daily Animal Checks

Each room has a daily care log on which animal care staff record the high and low temperature, high and low humidity, animal checks, and food and water checks. Each entry is initialed by the personnel performing the tasks. Every animal must be checked daily including weekends and holidays unless otherwise described in the IACUC protocol application and approved by the IACUC.

Animal Health Notification

Vivarium personnel or research staff noting any ill, injured, deceased, distressed, or otherwise abnormal animal must promptly notify the Animal Care Coordinator of the discovery. The investigator and Vivarium Director will both be notified of the issue via email and consulted regarding treatment. Documentation of the issue is created and maintained by the Animal Care Coordinator or the Vivarium Director on an Animal Health Report Form placed with the care log. Once the problem is resolved, the form is retained in the LAP office.

Summary of Animal Room Documentation Requirements

The following items must be documented either daily or when the activity is completed. All entries must be initialed by the personnel performing the task. These records must be kept for three years after the IACUC protocol has expired.

1. Daily (7 days a week including weekends and holidays)
   a. animal health check
   b. food and water adequacy
   c. temperature – high and low
   d. humidity – high and low
2. Twice Weekly
   a. check/change bedding
   b. change water bottle
   c. sweep/mop floors
3. Every Two Weeks
   a. change wire bar lid and microisolator top
4. Monthly (some species will be monthly; some tasks will be done monthly if needed)
   a. change brooms, mops, dustpans, feed containers, waste containers
   b. clean sinks and door jambs
   c. replace air filters, clean air vents
   d. sanitize room and drains
   e. change rack
Animal Use

Animal Numbers
All animals purchased, bred, or transferred from another project or otherwise acquired for use in research or teaching must be listed on an IACUC approved protocol prior to acquisition. There is an emergency holding protocol for unexpected circumstances when animals must be held on a valid protocol. Approved protocols list the total number of animals to be used and their numbers within each pain class. Animal usage is tracked against the approved number through animal deliveries, transfers, and numbers weaned. Animal transfers require permission and documentation with an Animal Transfer Request Form found on the IACUC website (even if transferred under the same investigator). An amendment requesting an increase in animal numbers should be sent to the IACUC for approval if an investigator needs additional animals.

Animal Acquisition and Ordering
Animal order forms are available on the IACUC website. Please submit the completed form to the LAP office for approval. Once the order request has been approved, the order will be placed, and the investigator will receive an order confirmation. If any discrepancies are noted with the confirmation, the investigator must contact the LAP office immediately upon discovery and prior to shipment of animals.

Animal Use Documentation
Care charges are billed by investigator account number. Charges are tabulated according to posted animal inventory logs (located outside each housing room). Investigators are responsible for accurate accounting of their animals. Mice are calculated per box, while all other animals are calculated individually. When weaning is performed, weanling numbers must be reported in the comment section of the animal inventory log. New individuals/cages must be added to the inventory log. Discrepancies noted will be added to inventory by care staff. Weanings and separations done by care staff will be noted on the inventory log by care staff. Please do not add extraneous information to this sheet. If you need to communicate about a concern, please contact the care staff.

Animal Movement
In an effort to prevent disease transmission and disruption of research, investigators must contact the Animal Care Coordinator for approval prior to moving animals between housing rooms. Animals brought into the facility without authorization will be quarantined or euthanized at the investigator’s expense.
Quarantine/Acclimation

For animals obtained from a commercial vendor, there is a three-day acclimation period prior to being used in studies. The acclimation period release date will be noted on the cage cards that the LAP provides at initial set up of the delivery.

For animals not acquired from a commercial vendor, a description of the housing facility and animal health surveillance program of the animal source must be provided to the Director of the LAP. This information should include:

- pathogens excluded
- methods of screening
- frequency of screening
- pathogen outbreak history
- current pathogens housed in the facility
- copies of the screening history for the room(s) of origin for the past year

Once these documents are evaluated, import permission may be granted or additional testing may be required prior to shipping.

Upon arrival, animals will be placed into quarantine. Immunocompetent animals may follow one of two courses to achieve release from quarantine. All fees associated with testing and quarantine will be the responsibility of the investigator.

Method One: Every animal will be tested directly for pathogens after one week of acclimation. Serology, fecal samples, and fur swabs will be collected and tested. Once all results are negative, the animals will be released from quarantine.

Method Two: Animals will be entered into quarantine but will be tested via dirty bedding sampling. This process will last for 8 weeks. Once testing is negative, the animals will be released from quarantine.

Immune compromised animals will only receive Method Two for release from quarantine.

Weaning

Litters of mice and rats over the age of 21 days are considered adults and should be weaned accordingly, unless specific exceptions have been approved in the IACUC protocol application. Weanlings should be separated by sex into cages of 5 or fewer animals independent of their weight at weaning. Care must be taken to ensure that the weanlings can reach both food and water.

Animal Procedures

Procedures such as blood draws, injections, behavioral testing, and surgeries performed on live animals must be described in the IACUC protocol application. The only exception is diagnostic sampling and medical care under the direction of the Vivarium Director for medical issues. Any stressful procedure, such as drawing blood, surgery, or euthanasia must be done outside of the animal housing rooms to avoid unnecessary stimulation of the other animals in residence. Procedure/surgery rooms are available for use by appointment.
Animal Health and Veterinary Services

Rodent Health Monitoring
LAP routinely screens animals, particularly those in breeding colonies or long-term studies, for rodent pathogens. Environmental sampling is used through sampling of dust and dirty bedding, as well as fecal testing and other non-sacrificial methods. These results help inform our biosecurity policies and determine order of room entry.

Reporting Ill, Injured, or Deceased Animals
If an ill, injured, or deceased animal is found during a daily health check, the animal must be reported to the Animal Care Coordinator, who will notify the Vivarium Director and the principal investigator by email. Cages will be flagged with a pink “Health Problems” flag or a blue “Death” flag behind the cage card. Documentation of the issue is created and maintained by the Animal Care Coordinator or the Vivarium Director on an Animal Health Report Form placed with the care log. Once the problem is resolved, the form is retained in the LAP office.

If the ill or injured animal requires immediate veterinary care, contact the Animal Care Coordinator at 618-453-6454. On holidays, weekends, or after-hours, call 618-713-1442. Less urgent cases may be reported via email for evaluation on the next business day. Veterinary staff will examine the animal and contact the investigator to discuss the recommended plan. If the investigator or designee cannot be reached, veterinary staff may proceed to treat or euthanize the animal as dictated by medical and ethical discretion. Current contact information must be available to LAP staff so that investigators and their staff can be consulted.

Euthanasia
The method of euthanasia must be described in the approved IACUC protocol prior to use. In the case of an emergency, an alternative method approved by the AVMA Guidelines for the Euthanasia of Animals: 2020 Edition may be used only under the direct guidance of the attending veterinarian.

CO2 Euthanasia of Rodents
Medical grade compressed CO2 gas in cylinders is the only allowable source of carbon dioxide. CO2 generated by other methods (e.g., dry ice) is unacceptable. CO2 delivery must be monitored by a flow
meter or preset system to ensure CO2 displaces mare at 30-70% of chamber volume per minute. Please contact the Animal Care Coordinator for assistance.

Rodents, especially neonates, are particularly resistant to euthanasia by overdose of inhaled agents such as CO2, or even injectable agents. For this reason, the AVMA Guidelines and the SIU IACUC require a secondary physical method of euthanasia for all rodents after the animal is euthanized and prior to carcass disposal. Acceptable secondary physical methods for adult and young rodents include:

- Decapitation
- Cardiac Perfusion
- Removal of vital organs (heart, lung, brain)
- Opening of the chest cavity to induce pneumothorax
- Exsanguination
- Cervical Dislocation

Note that none of these procedures may be performed in conscious animals without specific IACUC approval.

All persons conducting euthanasia must have received adequate training. This training must be documented and this documentation should be available for review by the IACUC.

Carcass Removal
If the animal's death was unexpected, it must be reported to the LAP as an abnormal event. The LAP Director may request a necropsy +/- testing to determine cause of death. In this case the carcass should be stored (not frozen) in the carcass refrigerator in the left hand outer hallway of LSIII. Dead animals and tissues must not be disposed of in the regular trash. If no further analysis of the carcass is anticipated, it should be disposed of in the carcass freezer. Properly bag and label all carcasses.

CEHS removes and properly disposes of freezer contents regularly. If you have any questions about proper disposal of chemicals or other waste, please contact CEHS for guidance at 618-453-7180.

Drugs, Chemicals, and Gases

Waste Gases
Any volatile chemicals or anesthetic gases must be scavenged by a biosafety cabinet, a fume hood or a portable carbon filter (F/Air type canisters if using an anesthetic vaporizer.) If you are uncertain of the device you want to use, contact CEHS for guidance at 618-453-7180.
Showers and Eyewash
The emergency shower and eyewash facilities are located in the short hallway on the right side of the vivarium. These are maintained in accordance with regulation.

Controlled Substances
Any investigator using controlled substances (Schedule I-V Drugs) in research must obtain their own DEA and state controlled substance licenses. It is a felony to provide a controlled substance to a person who is not registered with the DEA or who is not one of your authorized users (as declared to the DEA). Storage and use of controlled substances should meet or exceed DEA and State of Illinois Standards.

Expired Materials
The use of expired drugs, chemicals, disinfectants, and other substances on live animals is an unacceptable practice. Expired materials must be labeled as expired and stored separately from unexpired materials until proper disposal. Contact CEHS for guidance at 618-453-7180.

Reporting of Animal Welfare Concerns

Southern Illinois University will immediately investigate any allegations of suspected misuse of animals or deficiencies regarding the care and use of research or teaching animals. Any individual reporting a suspected deficiency need not identify themselves to make a report. If an individual chooses to identify themselves, their identity will be kept in strictest confidence.

To make a report or discuss a concern or perceived problem:

- Report online at: https://www.surveymonkey.com/r/siuiacuc22

- Contact the IACUC chairperson or any member of the IACUC. Each facility that houses animals must have a whistleblower poster prominently posted. These posters contain the names and contact information of the IACUC members. They also have a QR code to scan with a phone to report online immediately.

The federal Animal Welfare Act requires Southern Illinois University, as a registered research facility, to ensure that all scientists, research technicians, animal technicians and other personnel involved in animal care, treatment, and use are qualified to perform their duties. The SIU IACUC is charged with the responsibility of investigating complaints of misuse of animals used in instruction and research.