

**Northern Kentucky University
Institutional Animal Care and Use Policies**

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**Northern Kentucky University
Institutional Animal Care and Use Policy**

Mouse Husbandry & Breeding Colony Maintenance Policy

The Institutional Animal Care and Use Committee (IACUC) has established this policy regarding housing limits and breeding colony standards for mice. This policy applies to all NKU mouse users.

- 4 adult mice or 2 adult mice plus 1 litter are permitted per cage. Adult is defined as 30 days of age or older.
- For any litters, the Principal Investigator (PI) will tag the cage “TIME TO WEAN” when the litter is 21 days of age. It is the PI’s responsibility to separate the cage by 25 days of age.
- If breeding pairs are being maintained and a second litter is born, the older litter must be removed. The investigator has until the next business day to do so. The older litter may be housed in one cage and will then be subject to the weaning guidelines listed above.
- Harem breeding mice is defined as housing 1 male mouse and 2 or 3 female mice in one cage for breeding purposes. If harem breeding mice, only 2 adult mice and 1 litter may be housed in one cage. To prevent overcrowding, female mice must be housed individually or with one other mouse (male or non-pregnant female) when visibly pregnant. If the pregnant female mouse is housed with a male mouse, the housing is then subject to the breeding pair guidelines listed above.
- Refer to page 27 of the *Guide for the Care and Use of Laboratory Animals* (The Guide) for guidelines on the minimum standards for cage size. Copies of the Guide are available from the Office of Research, Grants & Contracts.

As stated on pages 26 and 27 in *Guide for the Care and Use of Laboratory Animals* (The Guide), “Depending on a variety of biologic and behavioral factors, group-housed animals might need less or more total space per animal than individually housed animals....An animal’s space needs are complex, and the consideration of only the animal’s body weight and surface area is insufficient. Therefore, the space recommendations presented [on page 27] are based on professional judgment and experience and should be considered as recommendations of appropriate cage size for animals under conditions commonly found in laboratory animal housing facilities.”

If any PI feels there is a scientific justification for an exception to this policy, a written request signed by the PI should be submitted to the IACUC Chairperson. Approval of the request for exception will be at the IACUC’s discretion. Any questions about this policy should be directed to the Office of Research, Grants & Contracts at X 5137.

**Northern Kentucky University
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Tail Clipping and Toe Clipping of Mice and Rats

For genotyping purposes or identification, mice and rats are often tail clipped or toe clipped. When either of these methods is necessary, the procedure must be described in the approved protocol and the guidelines in this policy adhered to unless the IACUC has approved an exception to this policy in the protocol. This policy does not apply to “nicking” the tail vein for blood collection.

Tail clipping on mice and rats without anesthesia can be performed only once on animals up to the time of weaning (approximately 21 days of age), removing no more than 0.5 cm of the tail. Anesthesia is required if the animal is over 21 days of age, more than 0.5 cm of the tail is removed, and/or when the procedure must be repeated.

According to *the Guide for the Care and Use of Laboratory Animals* (Guide), toe clipping as a method of identification should be used only when no other individual identification method is feasible and should be performed only on altricial neonates. Adequate scientific justification for this procedure must be provided in the protocol and approved by the IACUC. This method is only acceptable when the research is being performed on animals less than 7 days old, and the animals cannot be identified in any other manner.

**Northern Kentucky University
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Policy on the Use of Expired Drugs & Medical Supplies

The USDA's Animal Care Policy Manual states:

“The use of expired medical materials such as drugs, fluids, or sutures on regulated animals is not considered to be acceptable veterinary practice and does not constitute adequate veterinary care as required by the regulations promulgated under the Animal Welfare Act. All expired medical materials found in a licensed or registered facility are to be brought to the attention of the responsible official. The facility must either dispose of all such materials or segregate them in an appropriately labeled, physically separate location from non-expired medical materials.”

The NKU IACUC concurs that the use of expired drugs in research animals is an unacceptable threat to animal welfare and an unacceptable threat to the study's scientific validity; therefore, all research staff should check their inventory of drugs and supplies on a monthly basis. All expired items, except controlled substances, must be discarded. Controlled substances must be clearly labeled as expired and stored in a physically separate location from the rest of the stock until they can be discarded in a lawful manner. Expired supplies (e.g. suture materials) may only be used in acute terminal procedures. Expired supplies must be described in detail in the protocol and approved by the IACUC prior to use. Expired supplies to be used in acute terminal procedures must be labeled and stored in a physically separate location from non-expired medical material.

Any expired items not used in animals must be clearly marked “FOR IN-VITRO USE ONLY – NOT FOR USE IN ANIMALS.” If expired items are not clearly marked, the IACUC will assume it is being used in animals and act as stated in this policy. Any rooms listed on an approved protocol will be thoroughly inspected by the IACUC at least every 6 months. The IACUC inspection team may inspect all cabinets, drawers, and other storage units in the room.

Upon finding recurring deficiencies regarding this policy, the IACUC may request the PI to explain to the IACUC at a convened meeting why expired items were discovered in his/her laboratory and/or suspend the appropriate protocol(s).

Questions about this policy should be directed to the Office of Research, Grants & Contracts at X 5137.

**Northern Kentucky University
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Exceeding Animal Allotment Policy

This policy pertains to protocols with rodent breeding colonies. The IACUC understands that estimating the number of offspring produced under an animal protocol is, at best, guessing. If the PI underestimated the number of offspring produced, exceeding the approved allotted amount commonly occurs. The PI must keep track of his/her research animal populations and will notify the IACUC chairperson as he/she approaches his/her animal allotment. The IACUC Chairperson may approve a 10% increase of the approved allotment on behalf of the Committee. Although breeding rodents may continue, no animals may be purchased under or transferred to the protocol involved. The PI must submit a modification to increase the animal allotment within 30 calendar days after notifying the IACUC chairperson of the impending excess. The modification must justify the increase over the originally approved number.

**Northern Kentucky University
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Cage Identification Policy

The *Guide for the Care and Use of Laboratory Animals* recommends the following information be listed on cage cards:

- Investigator Name
- Protocol Number
- Species Strain/Stock
- Sex & Age or Weight
- Source
- Date of Birth or Arrival Date

NKU's IACUC requires that each cage be identified with the above referenced information, unless one of the following scenarios occurs:

- If all of the cages in the animal room are under one protocol, the room door may be labeled with the above-referenced information. Each cage does not require a cage card with this information unless this information differs (e.g. different dates of birth or dates of arrival).
- If all of the cages on a rack (one or both sides) are under one protocol, the rack may be labeled with the above-referenced information. The Principal Investigator is responsible for the cages placed on the rack. Each cage does not require a cage card with this information unless this information differs (e.g. different dates of birth or dates of arrival).

It is the Principal Investigator's responsibility to ensure that each cage can be clearly identified. All cages with the animals left unattended must be identified with the above-referenced information.

Sentinel animals are exempt from this policy.

**Northern Kentucky University
Institutional Animal Care and Use Policy**

Change or Addition of PI to an Approved Protocol

In cases where the Principal Investigator (PI) of an approved protocol is replaced with a new PI, the following steps will be taken:

- A new endorsement page must be signed by the new PI and the Department Chair. A fully signed copy must be forwarded to the IAUCU Chairperson
- IACUC Chairperson will function as the designated reviewer and evaluate the new PI's experience as relevant to the procedures in the protocol
- IF the new PI is deemed acceptable, the IACUC Chairperson will approve this action
- An approval letter, signed by the IACUC Chairperson, will be sent to:
 - The new PI
 - The Department Chair
 - The Research, Grants & Contracts Office

In cases where a second PI, or co-PI, is added to an approved protocol, the following steps will be taken:

- A new endorsement page must be signed by both PIs and the Department Chair
- The IACUC Chairperson will function as the designated reviewer and evaluate the additional PI's experience as relevant to the procedures in the protocol
- If the additional PI is deemed acceptable, the IACUC Chairperson will approve this action
- An approval letter, signed by the IACUC Chairperson, will be sent to:
 - Both PIs
 - The Department Chair
 - The Research, Grants & Contracts Office

The Office of Laboratory Animal Welfare (OLAW) identifies the changes listed above as significant changes that require IACUC review and approval. The University's IACUC has chosen to designate review of these changes by the IACUC Chairperson. Approval of these changes will be made at his/her discretion. The Committee will be notified of these changes during the next convened meeting.

**Northern Kentucky University
Institutional Animal Care and Use Policy**

**Policy on Requirement for Anesthesia of
Rodents for Retro-orbital Bleeding**

All rodent animals on the grounds of Northern Kentucky University or to be used on Northern Kentucky University approved animal use protocols must be anesthetized prior to the collection of blood by the retro-orbital bleeding technique. Investigators are reminded that they may be granted an exemption from this policy if they request it in writing, and provide valid scientific reasons why anesthesia will interfere with the proposed study results.

Current state-of-the-art veterinary medical practice is in agreement with the requirement to make anesthesia mandatory for rodent animals which are to undergo retro-orbital blood collection, as this procedure is considered inhumane unless the rodent animals are under surgical-level anesthesia.

**Northern Kentucky University
Institutional Animal Care and Use Policy**

Submission of Protocol Updates

To avoid protocols expiring with animals in-house, the IACUC strongly recommends the submission of protocol updates 2 months prior to the expiration of the protocol. This ensures that sufficient time is available for both the preliminary review and the IACUC review. It does not guarantee approval as changes or additions may be required.

Protocols are valid for no more than three years. The PI is responsible for updating the protocol yearly and submitting the renewal to the IACUC Chairperson.

Protocol updates should be submitted to the IACUC Chairperson for preliminary review. This process simply ensures the protocol is in adequate condition for IACUC review. It is preferred that protocols be submitted via email and the signature page mailed.

**Northern Kentucky University
Institutional Animal Care and Use Policy**

Aseptic Technique in Rodents

As stated in the *Guide for the Care and Use of Laboratory Animals* (Guide), appropriate aseptic technique is required for all survival surgeries in vertebrate animals. The requirement is fully described in the Guide, pages 60-64. Northern Kentucky University's animal care program must abide by the recommendations in the Guide, which is the book referenced by regulatory oversight agencies and the Association for the Assessment and Accreditation of Laboratory Animal Care (AAALAC).

Aseptic technique for rodents includes the following steps:

- A mask must be worn while performing surgery
- A lab coat or surgical scrubs must be worn during surgery (minimally a surgical scrub shirt may be worn); “street clothes” are not acceptable
- Sterile surgical gloves must be worn
- Surgical instruments must be sterilized prior to initial use
 - Ensure proper sterilization with autoclave tape or indicator strips
- In between animals, the instruments must be sterilized
 - Glass Bead Sterilization- instruments must be wiped or rinsed prior to submersion in the beads
 - Immersion in chemical sterilant – instruments must have proper contact time according to manufacturer recommendation, rinse instruments with sterile water or sterile saline after immersion before using in animals
- A portion of the lab must be dedicated for the surgery; other procedures may not be performed in this dedicated space during the surgery
- Animals must be surgically prepared as described in the approved IACUC protocol (e.g. shave hair, alternate betadine and alcohol scrub, etc.)

A sterile field must be maintained throughout surgery. A sterile surgical drape may be used to prevent contact with nonsterile surfaces and aid in maintaining a sterile field. Sterile instruments and gloves must not come into contact with any nonsterile surface (i.e. areas of the animal that have not been surgically prepared, nonsterile areas of the surgical space).

Exceptions to this policy or recommendations in the Guide must be detailed in the approved IACUC protocol. Scientific justification must be provided for such exceptions.

**Northern Kentucky University
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Labeling Requirements for Secondary Storage Containers

The Institutional Animal Care and Use Committee (IACUC) is responsible for inspecting areas where vertebrate animals are housed or utilized. The IACUC must ensure compliance with IACUC policies and federal regulations. In order to complete this, drugs and chemicals are inspected. Secondary containers in animal housing and procedure areas must be labeled according to this policy.

- Name of substance(s)
- Concentration of primary substance(s)
- Date of expiration per manufacturer or date the solution was placed in the secondary container

For in-vitro drugs and chemicals, the secondary container must be labeled with the name of the substance as required by OSHA. In-vitro substances should also be labeled and stored according to IACUC Policy #003.

**Northern Kentucky University
Institutional Animal Care and Use Policy**

Post Operative and Procedural Care Policy

Post-procedural care is a critical component of maintaining animal well being. Documentation of the clinical observations and treatments administered to certain animals (excluding rodents) recovering from anesthesia is required and reviewed by the USDA and AAALAC.

Recovery from anesthesia/surgery has three component time periods requiring different levels of monitoring.

1. Anesthesia Recovery Period: This is the period until the animal is extubated, if applicable, and able to maintain itself in sternal recumbancy (i.e. autonomously maintain normal upright posture) in a seated position.
2. Acute Post-procedural Care: This is the period of time after the animal can maintain sternal recumbancy and is able to get up and freely move about without staggering. This period may last from an hour to several hours depending on the anesthetic agent used.
3. Long Term Post-procedural Care: This is the period following recovery until the skin has healed (usually 10-14 days after surgery). Abnormal or unexpected responses having the potential for negative impact on the health of the animal (i.e. adverse clinical findings) observed during any of these recovery periods must be reported immediately to the Principal Investigator.

Emergency veterinary service is available during or after business hours by calling Dr. John Sector's office and pager at 859-371-4890.

Monitoring and Record Keeping Requirements:

All entries in the animal records are to be initialed so the clinical veterinary staff, USDA inspector, and AAALAC site visitors can determine whom to contact if they have questions. Any medications administered must be recorded throughout the recovery periods. Records are to be concurrently maintained and easily accessible.

Surgical and/or procedural records must contain the following: date, protocol number, Principal Investigator, species, animal identification number if a USDA-regulated species, pre-procedural health assessment, anesthetics and analgesics administered (dosage and route), description of procedure (procedure start and stop times), assessment of procedure, and post-procedural care. Once the procedure is completed, the animal's cage card or associated card must reflect the date and type of the procedure performed.

All animals must be monitored for signs of pain or distress during the post-procedural period. Those procedures that are likely to produce pain in humans are to be considered painful in animals. Signs of pain include grinding of teeth, licking or guarding of the painful area, vocalization, restlessness, lack of mobility, refusal to groom, abnormal posture, failure to show

normal patterns of inquisitiveness, and failure to eat or drink. Pain relieving drugs are required when the animals are perceived to be experiencing pain or distress unless prior approval to withhold these drugs has been obtained by the IACUC. The drugs must be either approved in the IACUC protocol or prescribed by the veterinary staff.

Guidelines for non-rodent mammals

Anesthesia Recovery Period:

Observations must be recorded at intervals of 20 minutes until the animal is ambulatory. The parameters to be observed and recorded include heart rate, respiratory rate, mucus membrane color capillary refill time, body temperature and any abnormal findings. The animal must be rotated or turned over every 30-60 minutes to facilitate respiration and avoid dependent edema.

Acute Post-procedural Care Period:

Animals are to be monitored and observations are to be recorded. The animal's ability to ambulate normally and behavioral signs of pain must be assessed and recorded.

Long Term Post-procedural Care Period:

The animals are to be observed at least daily and any abnormal observations of the animal or surgical site must be reported to the veterinary staff or PI immediately.

Guidelines for Rodents:

Anesthesia Recovery Period:

Observations are made at intervals of no more than 20 minutes until the rodent is ambulatory. Observations need not be recorded unless abnormal findings are found. The parameters to be observed include respiratory rate and mucus membrane color. The animal must be rotated or turned over every 30-60 minutes to facilitate respiration and avoid dependent edema.

Acute Post-procedural Care Period:

The animal's ability to ambulate normally and behavioral signs of pain must be assessed and recorded.

Long Term Post-procedural Care Period:

The animals are to be observed at least daily and any abnormal observations of the animal or surgical site must be reported to the PI immediately.

**Northern Kentucky University
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Specifications for Satellite Animal Procedure Areas

Security:

Animals must be attended by an approved animal user for that protocol (i.e. approved animal user in the area) or held in a locked laboratory. As stated in the *Guide for the Care and Use of Laboratory Animals*, facilities should have security features such as card-key systems or alarms.

Cleanliness:

The area must be kept clean (i.e. free of dirt, blood, bedding, etc.) if being used on an ongoing basis. Procedure areas must be wiped down with an antimicrobial agent before and after animal use to minimize allergens and possible exposure to microorganisms/infectious agents. More stringent requirements are required for surgery. Please reference OLAW's CD-ROM "Training in Survival Rodent Surgery," which may be obtained through the Office of Research, Grants & Contracts.

Occupational Health and Safety:

All personnel exposed to animals in the procedure area must be covered under the University's Occupational Health and Safety Program.

Justification:

Justification (for use of area – not procedures) must be provided to perform protocol procedures on live animals outside of core animal housing facilities. It must be based on necessity for the science (i.e. equipment cannot be secured, transported or stored on site; animals must be continuously monitored as part of the study, etc.).

Returning Animals:

Animals must be returned to core animal housing facilities in accordance with the IACUC protocol and IACUC Policy on Housing Animals in Laboratories. All dirty cages must be returned to the core animal housing facilities within 24 hours to minimize researchers' exposure to allergens.

Suggestions:

If chemical or gas anesthetics are going to be used, it is recommended that a chemical fume hood be available in the room

A space for secured compressed gas tanks, such as CO₂

Sealed benches that can be sanitized

A sink with hot and cold running water be available in the room

If controlled substances are used in the room, a secured lockable area should be available (e.g. locked cabinet, mounted lock box)

NKU IACUC Policy #012

Investigators must review IACUC Policy regarding Housing Animals in Laboratories to ensure the procedures being performed in the room cannot be defined as housing (animals held in the satellite area overnight with the exception of USDA covered species, which is over 12 hours).

**Northern Kentucky University
Institutional Animal Care and Use Policy**

**Requirement for Safety Office Approval
Prior to IACUC Protocol Approval**

In order to meet accreditation requirements, the IACUC will determine safety risks and request rulings from appropriate safety committees or offices (i.e. Physical Plant, Environmental Health & Safety). While IACUC protocols will be reviewed prior to approval by the safety committees, IACUC approval will be withheld until the appropriate safety office(s) approval is obtained. Ordering of animals will not be allowed and animals in-house may not be used as well.

For protocol modifications, the modified portion of the protocol cannot be approved until the necessary safety office has approved it.

Principal Investigators are strongly advised to check with appropriate safety offices prior to the submission of animal protocols to the IACUC.

Contact information for all University safety offices can be found on the IACUC link on the RGC website at rgc.nku.edu/.