Ohio University IACUC Approved Guidelines
Revised & Approved: May 4, 2020

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A. Ohio University Use of Controlled Substances in Research

Objective

The objective of the Controlled Substances Policy is to ensure compliance with state and federal regulations governing the use of Drug Enforcement Agency (DEA) controlled substances.

I. Policy

Ohio University requires that all individuals working with DEA controlled substances be registered with the DEA and with the Ohio Pharmacy Board and comply with state and federal regulations regarding the acquisition, storage, use and disposal of those substances.

II. Responsibilities

Each principle investigator conducting research activity using Drug Enforcement Agency (DEA) controlled substances will be responsible for registering with the DEA and with the Ohio Pharmacy Board and for assuring compliance with applicable state and federal regulations. The Vice President for Research will be responsible for assisting principle investigators in complying with applicable rules and regulations. This will include educating researchers about the requirements, assisting them as necessary during implementation, and providing regular oversight to ensure compliance is being maintained.

III. General Information and Procedures

Both the state and federal law classify controlled substances into five categories according to their potential hazard. Schedule I substances are the most hazardous, schedule V the least hazardous. Every person conducting research activity using a controlled substance is required to register with the Drug Enforcement Agency (21 CFR, 1301.21). Registration is required annually and there is no fee for state employees.

For substances in Schedule II-V, one should submit DEA Form 225. For Schedule I substances, in addition to DEA Form 225, the applicant is required to submit 3 copies of a research protocol as defined in 21 CFR, 1301.33.

The registrant is responsible for managing the controlled substances in accordance with the requirements of the regulations, including inventory, record keeping and security provisions and disposal.
The Ohio University IACUC will uphold the Ohio University policy on the use of controlled substances that are used in live vertebrate animal research.

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B. Ohio University Endpoints Guidelines for Experimental Live Vertebrate Animals

Compliance with regulatory guidelines requires that death as an endpoint to experimental processes be fully justified and documented. Most experiments do not require death as the endpoint for the study and euthanasia should be performed on all moribund experimental animals unless the justification for death as an endpoint is included in the approved protocol. The following guidelines will allow consistent assessment of animals to determine the endpoint for experimental manipulations not requiring death as an endpoint.

Qualification Criteria for Automatic Euthanasia Without Prior Investigator Notification

The following condition will automatically qualify an animal for euthanasia upon notification of the Laboratory Animal Resources (LAR) Director and/or Attending Veterinarian, unless an approved exception (protocol) is on file: Lack of consciousness with no response to external stimulus.

Emergency Euthanasia

LAR technicians may also request emergency euthanasia for animals in extreme circumstances. Animals will be evaluated by the LAR Director and/or the Attending Veterinarian, and a decision on immediate euthanasia will be made at that time. An attempt will be made to notify the investigator(s) prior to emergency euthanasia if at all possible.

Criteria for Entry into the Endpoint Evaluation Program

Animals in a moribund state, animals with tumors, and animals with significant health problems will be entered into the endpoint evaluation program for daily assessment of their conditions unless an approved exception (protocol) is on file.

The following conditions will automatically enter the animal into the endpoint evaluation program:

1. Inability to maintain normal, age-appropriate posture
2. Inability to reach food and/or water
3. Chronic diarrhea (persisting for more than 7 days)
4. Labored breathing
5. Anorexia (failure to eat for longer than 48 hours in mammals, non-mammalians qualify if failure to eat persists longer than twice their normal feeding intervals)
6. Dehydration
7. Tumors (per the guideline below), unless an approved exception (protocol) is on file
8. Skin lesions
9. Eye Problems
10. Prolapsed uterus/rectum
11. Persistent lack of urine/fecal output

Any of the above conditions qualify the animal for euthanasia. The final decision on euthanasia is made by the Attending Veterinarian and/or LAR Director.

**Investigator Notification**

Investigators will be given a twenty-four hour notice to collect tissues or otherwise use the animal prior to its disposal, if the animal is not in extreme pain and distress. After this 24 hour period, euthanasia for animals meeting the above criteria will be carried out by the LAR technicians, automatically, with no further warning.

**Monitoring and Record Keeping**

Animals being monitored for endpoint status will be evaluated by a LAR technician at least once each day, including weekends and holidays. A written record of the observations will be kept on either an observation chart or the cage card. The observation record will be forwarded to the Attending Veterinarian and the Investigator when the animal is either euthanized or removed from the endpoint evaluation program.

**Tumor Guidelines**

The following guidelines apply to animals with tumors:

1. Tumors should not exceed 10% of the animal’s body weight. Animals in which tumors are expected will be weighed prior to the beginning of the experiment and that weight used as a baseline to measure tumor weight. Animals exhibiting this condition will be immediately euthanized.
2. Tumors should not interfere with the animals’ ability to move about the cage, or to reach food and water. Animals may be offered food and water on the floor of the cage if they can still move about the cage but not reach the elevated food and water containers. Animals unable to eat and drink will be immediately euthanized.
3. Ulceration of tumors is a criteria for euthanasia unless an approved exception (protocol) is on file.

The final decision on euthanasia is made by the Attending Veterinarian and/or LAR Director.
C. Ohio University Live Vertebrate Definition

Mammals:

The IACUC will consider the fetuses of mammals “Live Animals” at the live birth of such animal(s).

Non-Mammalian Vertebrates:

The IACUC will consider non-mammalian vertebrates (i.e., reptiles, amphibians, fishes, birds) “Live Animals” at the time the species requires an external food source or are no longer confined to an amniotic egg.

D. Ohio University Use of Non-Pharmaceutical Grade Compounds in Live Vertebrate Animals

Pharmaceutical grade medications are standard for use, even in acute procedures, when they are available. Non-pharmaceutical grade chemical compounds should only be used after review and approval by the IACUC. Justification must be provided for use, including scientific necessity and/or lack of availability for a pharmaceutical-grade medication. For more specifics, see the Guide, Eighth Edition, page 31.

E. Ohio University Institutional Animal Care and Use Committee Protocol Review Procedures for Purchase of Custom Antibodies

Review of protocols requesting purchase of custom antibodies, where the antibody production does not involve use of live vertebrate animals at Ohio University, will be reviewed by the Chair of the Institutional Animal Care and Use Committee to assure they meet the requirements; the Chair will decide approval. This applies only to antibodies produced specifically for an investigator and does not apply to antibodies which are available commercially for purchase. This is necessary because, even if custom antibodies are produced outside Ohio University by a vendor, the investigator may be required by granting agencies, such as NIH, to have the work reviewed by the IACUC. Failure to comply with this guideline can result in delay or denial of your grant by the granting agency.
If you are purchasing custom-made antibodies from commercial vendors, you must provide the following requirements for review:

A. Vendor’s name
B. Vendor's OLAW “Assurance” number or assurance that the vendor’s IACUC has reviewed and approved the methods used to produce the antibodies
C. Vendor's USDA "Registration" number, if available.

F. Ohio University Rodent and Non-mammalian Aseptic Surgery Guidelines

1. A clean work area with a surface that can be sanitized must be used. The surgical area should be separate from the location used for anesthesia and hair removal. This site should be in a low traffic area and dedicated to surgery when in use as a surgical suite. It is preferable to locate the surgical suite away from windows, fans, fume hoods or vents, which can blow dust into the area. Consideration to maintaining the body temperature of the animal should be given.

2. The animal must be appropriately prepared, including removal of the hair for rodents and application of a disinfectant surgical scrub on the skin at the surgical site. The disinfectant should be in contact with the skin long enough to be effective; three consecutive scrubs using clean pads with each scrub are generally considered standard. Take care not to saturate the body with disinfectant since this can cause hypothermia. Examples of appropriate skin disinfectants include povidone-iodine and Chlorhexidine.

3. The use of sterile instruments and supplies is required. A sterilization procedure for instruments includes autoclaving, glass bead, gas sterilization or cold chemical sterilization. It is essential when using cold sterilization that only chemicals classified as “sterilants” be used. Note that the length of time for sterilization can be as high as ten hours with these products.

4. Serial rodent and non-mammalian procedures allow for use of instruments on more than one animal but care must be taken to prevent contamination between animals. At least two sets of instruments should be sterilized before beginning. Instruments should be wiped clean with 70% alcohol, sterile water or saline, and kept on sterile non-porous drapes between procedures. If contamination has occurred the instruments must be sterilized using a chemical agent or glass bead sterilizer for the length of time necessary for the method to be effective or replaced with a new sterile instrument pack. Use of two or more sterile packs can reduce the amount of down time if re-sterilization is needed. Sterilization of the instruments must occur before the next set of serial surgeries begins. If chemical agents are used instruments must be rinsed with sterile saline or water before being used on the next animal. It is recommended that instrument packs be used on no more than five animals before being re-sterilized.
5. Surgeons should wash their hands with an antiseptic surgical scrub preparation prior to beginning surgery. Sterile gloves, a mask, and a clean scrub shirt or lab coat are required to be worn by the surgeon and any assistants working in the immediate surgical field. New sterile gloves should be worn for each additional animal. If precautions are taken to minimize contamination of the surgical gloves it may be adequate to rinse the gloves with a disinfectant solution between animals. Care must be taken that no handling of non-sterilized items has occurred, or they must be replaced immediately with sterile gloves.

6. Postoperative care includes monitoring for infections, monitoring of the incision sites, and timely removal of skin sutures, clips or staples. A description of the appropriate postoperative care must be included in the protocol description.

7. In non-survival surgeries it may not be necessary to follow all techniques listed; however, at a minimum, the surgical site should have the hair removed, the surgeon should wear gloves, and the instruments and surrounding areas should be clean.

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G. Ohio University Mouse Tail Biopsy Policy

The IACUC encourages the use of the least invasive method of tissue sampling, e.g., ear punching. However, if tail biopsies are to be used, they must conform as follows:

**Procedure:**
- Sampling may be conducted no more than twice and if re-sampling is needed from the same mouse, no more than 5 mm in total of the distal tail should be harvested. In this situation, other tissue sources should be considered for harvest (e.g., ear pinna). Biopsy of tail tissue can be performed without general anesthesia or local analgesic in mice up to 28 days of age. General anesthesia or local analgesic is required when tail biopsy is performed on animals older than 28 days of age.
- Any bleeding at the tail tip must be controlled (hemostasis) following the biopsy. If less than 2 mm is taken, hemostasis can usually be achieved by direct manual pressure with clean paper towel or gauze on the end of the tail. If direct pressure does not stop the bleeding, the use of hemostatic agents [e.g., styptic powder (Kwik-Stop ®)] is recommended and should be readily available as a precautionary measure.

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H. Ohio University Murine Ulcerative Dermatitis (MUD)
Some laboratory mouse strains [e.g., C57BL/6J (B6)], are predisposed to developing severe ulcerative dermatitis. This has been well documented in the literature and is currently considered idiopathic in some strains. This condition can progress quickly and cause lesions over large parts of the body. Areas primarily affected are the neck, head and axillary regions of the limbs. This condition does not typically respond well to treatment and can become debilitating, if not addressed.

**POLICY:**
Laboratory Animal Resources (LAR) staff or any other caretakers that detect the beginning stages of ulcerative dermatitis are required to document the presence of the condition on the cage card, and to alert the protocol Primary Investigator (PI) of the malady. The PI is responsible for assuring that the following policy is adhered to, including any necessary treatments.

**EARLY TREATMENT MURINE ULCERATIVE DERMATITIS:**
Upon diagnosis, all mice exhibiting early symptoms of ulcerative dermatitis must be placed on a treatment program immediately (and the treatment recorded on the cage card) and the Attending Veterinarian must be notified. Alternatively, project PI may elect to euthanize the mouse at this time.

In mice that exhibit patches of ulcerative dermatitis with total area on the body less than the area of a 10 mm circle must:
1. Have their rear toenails trimmed immediately and at least every 10 days thereafter, &,
2. Be treated with one of the following options:
   - **Option A:** Treat topically with a combination antimicrobial and corticosteroid ointment as recommended by the Attending Veterinarian.
   - **OR**
   - **Option B:** Treat topically with antibiotic ointment with a topical pain reliever (as recommended by the Attending Veterinarian) twice a day for one week, then 3 times per week, for a duration determined by the Attending Veterinarian, following initial treatment.

**MANDATORY EUTHANASIA:**
As the ulcerative dermatitis progresses mice must be euthanized if any of the following apply:
1. There is a single ulcerative lesion greater than 10 mm in diameter, or
2. There are multiple ulcerative lesions that total 10 mm in diameter, or
3. There are any ulcerative lesions exhibited with additional signs of morbidity.
I. Ohio University Automatic (Instant) Designated Review

Protocols that fit within the following parameters may be reviewed by designated review without notification of the entire IACUC prior to the next convened meeting:

1. Addenda to currently approved protocols that include:
   a. Changes in Co-Investigators that do not require surgical or anesthetic expertise.
   b. Procedure site changes as long as they are within currently accepted areas.
   c. Animal number increases that do not exceed 10% of the original request excepting for species covered by USDA, in which case any increase must be reviewed by full committee or designated member review.
   d. Additions or changes in stocks, strains or breeds of animals excepting for genetically engineered stocks, strains or breeds.
   e. Minor non-anesthetic, non-analgesic, non-surgical changes in procedures that do not impact animal welfare.

2. Protocol submissions for approval that are:
   a. For commercially produced custom antibodies with no live animal use on site (Ohio University). The vendor must have a current PHS assurance and, where applicable, a current USDA license and an approved protocol in place. The protocol submission to Ohio University is used to assure that the appropriate assurances, licenses and approved protocols are in place at the vendor site.
   b. Noninvasive, observational only field studies in which no capturing, banding, bleeding or other handling of animals occurs.
   c. Noninvasive use of animals for film/media/art.

J. Ohio University Training and Continuing Education Requirements

Animal Users:
In accordance with Ohio University’s PHS Assurance, all persons handling live vertebrate animals and/or listed on an IACUC protocol are required to complete the Collaborative Institutional Training Initiative (CITI) online animal user course, which is entitled “Investigators, Staff and Students” in the Working with the IACUC curriculum, prior to protocol approval and animal use. Once the initial course is completed and passed, each person handling live vertebrate animals is required to complete the refresher course, which is located on the CITI site, once every three years.
It is recommended that all persons using live vertebrate animals also complete the species and procedure-specific courses available on CITI that are relevant to their work. In addition, these persons should be given sufficient training by the primary investigator (PI) to successfully accomplish the work assigned with live vertebrate animals. This training must be documented by the PI and a copy made available upon request of the IACUC.

Following is a format suggested for the training log.

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<tr>
<th>Date</th>
<th>Trainee</th>
<th>Trainer</th>
<th>Procedure Being Taught</th>
<th>Proficiency Demonstrated</th>
<th>Initials of Trainee</th>
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Personnel in laboratories who do not have involvement in handling of live vertebrate animals are not required to be listed on the protocol and, if not listed, do not need to complete animal use training. However, all personnel that are listed on the protocol must complete CITI training even if they do not handle live vertebrate animals.

**Classroom Use:**
Students who will only have transitory contact with live vertebrate animals during the completion of coursework where that contact occurs exclusively in the classroom setting and is supervised by an instructor who has completed the required CITI training are not required to complete the CITI course. Instructors must provide a basic orientation to the animal use that will occur and have a written summary of that orientation available for review by the IACUC upon request.

**IACUC Members:**
Members of the Institutional Animal Care and Use Committee (IACUC) must complete the CITI course “Essentials for IACUC Members” in the Working with the IACUC curriculum.

**Laboratory Animal Resources (LAR) Staff:**
The staff members are required to complete the CITI course “Investigators, Staff and Students” in the Working with the IACUC curriculum, and to complete the refresher course once every three years. The LAR animal care staff is also required to complete
AALAS certification. Continuing education of the LAR animal care staff must be documented and these records made available to the IACUC for review upon request.

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K. Ohio University Designated Member Review (DMR) Subsequent to Full Committee Review (FCR)

This guideline is required to be reviewed and approved by all voting members at a convened meeting of the IACUC. Any changes must also be reviewed and approved by all voting members at a convened meeting of the IACUC.

This guideline allows the IACUC to decide by unanimous vote of a quorum of members present at a convened meeting to use DMR subsequent to FCR when modification is needed to secure approval.

Any member may, at any time, request to see the revised protocol and/or request FCR of the protocol.

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L. Ohio University Non-Significant Amendments

Amendments that fit within the following parameters are considered non-significant and do not require review by the IACUC. These amendments may be reviewed and approved by either a member of the Office of Research Compliance or by an IACUC member.

1. Changes in personnel other than Primary Investigator and Co-Investigators as long as the personnel are not performing surgery, anesthesia, or euthanasia. Added personnel must still meet training requirements.
2. Changes in procedure sites as long as they are within currently accepted animal use areas.
3. Changes in strain or breed which are not associated with use of a transgenic or knockout animal.
4. An increase in numbers of non-mammalian species, purpose bred mice and purpose bred rats of less than 10%.
5. Changes to titles
6. Changes to funding sources
7. Grammatical corrections for clarity
M. Ohio University Use of Expired Pharmaceuticals, Biologics, and Supplies in Live Vertebrate Animals

The use of expired pharmaceuticals and biologicals is not acceptable for use in live vertebrate animals. In particular, anesthetic, analgesic or euthanasia agents should not be used beyond their expiration date even in terminal procedures.

Use of other expired materials must be approved by the IACUC. Such approval must be contingent upon verification of efficacy beyond the expiration date, either by the manufacturer or the investigator if he or she can document efficacy in a way that is accepted by the IACUC. The use of the expired material must not impact animal welfare or the validity of the study in order for the IACUC to approve such use.

In the case of materials directly used with the animals that do not have expiration dates associated with them the investigator should clearly mark the date the material was purchased or produced to allow for evaluation by the IACUC on an ongoing basis.

N. Ohio University Social Housing of Laboratory Animals

Purpose:
The policy is to provide guidance for providing social housing to laboratory animals maintained at Ohio University under an IACUC approved protocol.

Background: The Guide for the Care and Use of Laboratory Animals (NRC, 2011) states:

“Appropriate social interactions among members of the same species (conspecifics) are essential to normal development and well-being (Bayne et al. 1995; Hall 1998; Novak et al. 2006). When selecting a suitable social environment, attention should be given to whether the animals are naturally territorial or communal and whether they should be housed singly, in pairs, or in groups. An understanding of species-typical natural social behavior (e.g., natural social composition, population density, ability to disperse, familiarity, and social ranking) is key to successful social housing.”

“Single housing of social species should be the exception and justified based on experimental requirements or veterinary-related concerns about animal well-being. In these cases, it should be limited to the minimum period necessary, and where possible, visual, auditory, olfactory, and tactile contact with compatible conspecifics should be provided. In the absence of other animals, enrichment should be offered such as positive interaction with the animal care staff and additional enrichment items or addition of a
POLICY:

Social housing of all social species of animals is the default housing environment at Ohio University. Single housing of social animals must be scientifically justified based on experimental design requirements in the Housing Data section of the LEO IACUC protocol form.

Social animals may also be individually housed due to the following factors:

* Incompatible animals with no other compatible conspecifics may be individually housed.
* Animals may be individually housed due to veterinary necessity
  - Entry or exit quarantine
  - Injury/disease treatment
  - Post-operative recovery
  - Breeding/parturition necessity
* Animals may be temporarily housed for procedural or experimental purposes.

In any of these cases, single housing of social animals should be limited to the minimum period necessary and, where possible, visual, auditory, olfactory and tactile contact with compatible conspecifics should be provided. In the absence of other animals, supplemental enrichment should be considered. Protocols experiencing an unusually high number of the exceptions listed above may require further IACUC oversight/approval. Moreover, single housing of social animals must be justified in the protocol and approved by the IACUC.

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O. Ohio University Animal Holding

Purpose:
This policy is to provide guidance for providing housing and care to laboratory animals maintained at Ohio University that, due to unforeseen circumstances, are not covered under an approved IACUC protocol.

Background: The Guide for the Care and Use of Laboratory Animals (NRC, 2011), PHS Policy, the USDA Animal Welfare Act and Ohio University Policy 19.049 all require IACUC oversight and approval of the use of live vertebrate animals in research and teaching. However, in very rare circumstances (such as IACUC suspension or expiration of an approved protocol), animals may need to be housed and cared for in
Ohio University animal facilities by Laboratory Animal Resources and overseen by the Attending Veterinarian.

POLICY:

Laboratory animals will typically not be ordered or received by Laboratory Animal Resources (LAR) without an IACUC approved protocol in place. In extraordinary circumstances animals may be ordered and received with the approval of the LAR Director and the IACUC Chair, if approval of an IACUC protocol is in process. In the instance of a protocol suspension, investigation or expiration, the IACUC will notify LAR immediately to initiate implementation of this policy.

- Any animal housed in Ohio University facilities and used in research, teaching, or testing that is NOT covered by an approved IACUC animal use protocol will immediately be subject to this policy.

- Animal rooms subject to this policy will be locked and signage will be posted indicating that the animals involved fall subject to this IACUC policy, and no research may be done or data collected from the animals.

- Once this policy is in effect, neither the Principal Investigator nor his/her staff will have access to the animals. No research, teaching, or testing may occur with animals which are subject to this policy. This includes data collection, observation, surgery, administering test substances, or any other experimental procedure. Medications needed for the welfare of the animal may be administered by order of the Attending Veterinarian at his discretion.

- All vertebrate animals subject to this policy will be cared for and maintained by LAR. Animals will be provided food, water, and housing in a manner appropriate for the species and within the guidelines of the Ohio University Animal Care and Use Program. Special husbandry procedures or concerns may be utilized, if necessary, for the welfare of the animals and if approved by the IACUC Chair, LAR Director, and the Attending Veterinarian.

PHS funding cannot be used to support the care and oversight of animals subject to this policy.

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P. Potential Conflict of Interest and Authority of the Chair

When the Chair of the IACUC believes that there is a potential Conflict of Interest (COI) for any reason whatsoever, such as when a protocol or amendment is submitted that includes the Chair of the IACUC, then the following process will be implemented.

a. The Chair will contact the Office of Research Compliance to notify the Director of
the situation and recuse themselves from the situation.
b. The Attending Veterinarian (AV) will be notified of the situation.
c. The AV will then serve as the acting Chair and determine the best course of action to address the item.
d. Once the issue is resolved then the AV will relinquish the duties of the Chair and the IACUC Chair will once again assume the duties as Chair.

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Q. Animal Facility Access

Satisfactory completion of the CITI basic animal use course “Investigators, Staff and Students” under the “Working with the IACUC Course” is required for access approval to the animal holding facilities. Primary Investigators requesting animal facility access for students or research staff not listed on an approved IACUC protocol will need to provide a CITI training certificate, as well as assurance that the person has been given the opportunity to be enrolled in the occupational health program by completing the Health and Risk Assessment form: https://www.ohio.edu/riskandsafety/docs/OccupationalHealthForm_AnimalResearch.pdf

Requests for animal facility access must be made by the Primary Investigator of the research project to the Director of Laboratory Animal Resources. These access requests must include the currently approved IACUC protocol number(s) that the requested person will be working on, along with their research role, university status and PID number (and/or assigned Proximity card number if applicable).

Access to the animal facilities by other outside visitors follows OU Policy 19.057. https://www.ohio.edu/policy/19-057.html

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R. Ohio University Animal Ordering and Tracking

Objective

The objective of the Animal Ordering and Tracking policy is to ensure that all animal transfers are handled in accordance with Ohio University requirements. This will help ensure that the health of the animals is maintained and the location of all animals is easily tracked.

All live vertebrate animals used for research, teaching, or testing at Ohio University must be procured through Laboratory Animal Resources. This includes both wild caught and domestic animals. All animals that will be housed in LAR animal research facilities must be imported into the facilities under the supervision, assistance, and approval of the LAR Director and Attending Veterinarian. An active, approved IACUC protocol must be in place to procure commercially produced animals through LAR, or to import animals from
other sources. Animals purchased from commercial vendors, and animals transferred from other institutions will be handled using the same tracking process.

All animals produced within Ohio University breeding colonies will be tracked by the Primary Investigator (or PI’s staff) of the IACUC protocol being used. Animals will be counted within one week of birth and added to the official protocol census sheets maintained on the animal facility doors.

All animals transferred onto an IACUC approved protocol from another OU IACUC protocol will be tracked by the primary investigator (or PI’s staff). Animals will be added to the official protocol census sheets maintained on the animal facility doors. It will be the responsibility of the PI (or PIs if different investigators hold the protocols) to ensure that the protocol census sheets are marked correctly with the animals being logged off of one protocol and onto another.

All animals transferred to other institutions must be exported under the supervision, assistance, and approval of the LAR Director and Attending Veterinarian. Health status, veterinary reports, and LAR facility information must be transferred to the other institution by the LAR Director or the Attending Veterinarian. Additional requirements, such as Material Transfer Agreements may need to be completed before a transfer is approved.