



Animal Program Policy

Title: Training of New Staff
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All staff planning to work with vertebrate animals must first complete all standard Forsyth onboarding, including New Personnel Safety Training with the Director of Occupational Health and Safety. Then, the PI or personnel must contact the IACUC Coordinator to request training. All steps below must be completed before gaining entry into the animal facility. The IACUC Coordinator will verify training dates and provide key card access once all requirements have been satisfied. Training required to gain entry into the animal facility consists of:

1. **Training Request Form to be completed and signed by both the personnel and the PI.**
2. **Personnel Amendment to be submitted by the PI – please note that the personnel must be assigned to work on a specific protocol(s) to begin the training process.**
3. **Occupational Health Questionnaire via the Forsyth intranet.**
4. **CITI online training.**
5. **CORI Acknowledgement Form to be submitted to Forsyth Human Resources.**
6. **Animal training provided by the Director of the Animal Facility.**
 - a. **Classroom training**
 - b. **Facility-wide training**
 - c. **Procedure-specific training**
7. **Read and understand the protocol(s) the individual will work on.**

On completion of the required steps above, personnel will be granted access to the animal facility via personal key card. It is unacceptable for personnel to share cards or open doors to give facility access to other personnel (AKA “piggybacking”) who have not satisfied all requirements for clearance.

Animal Training:

Irrespective of previous experience, all staff has to undergo this training to familiarize themselves with the lay out, set up and the management of the Forsyth Animal Program. The training is carried out in two stages. Power point presentation and walk through the Animal Facility.

A. Power point presentation:

Initially, all the staff due to undergo training are shown a power point presentation with detailed explanation of the training material. During this session, additional discussion and interaction is encouraged. Emphasis is made on important areas that are directly related to the welfare of the animals and what guidelines are strictly adhered to. Trainees are guided to clearly distinguish what they can do and what they cannot do, while working in the animal facility. The trainees are requested to fill an Animal Training Form and also indicate any past experience and whether they will need any special training in the following areas: Administration of injections (IM, IP or S/C), blood collection, identification with ear tags, collection of tissues, and tail snips or any other areas that need specific input.

Specific Items to be discussed include:

- **Role of IACUC** and the Forsyth Animal Care Program.
- **Introduction to IACUC Website**
 - IACUC Policies
 - IACUC Protocol Forms
 - Calendars to sign up for procedure rooms
 - Other documents on the website
- **Protocols:** Animal work cannot be carried out without an approved protocol. Names of the staff should be included in the protocols. All staff should familiarize themselves with the details of the approved protocols, their role and ensure that only approved procedures as indicated in the protocol may be carried out. Preplanning and scheduling is essential. Discuss any needs with the animal facility staff. (Yoga can ask specific questions about the relevant protocol at this time.)
- **Outside agencies:** AAALAC, OLAW (NIH), USDA, and the City of Cambridge oversee our program, including unannounced inspections.
- **Obtaining Animals:**
 - Animals can be obtained from approved vendors or from other institutions. If it is from approved vendors, contact the director for approval giving the protocol number and other details through the MIP system. Make sure to fill the form completely. (Protocol number required for approval) Animal numbers to be used are limited by the protocol. Animals from approved suppliers can directly go into the holding rooms.
 - If the animals are from other institutions, contact the director with details. Importation is approved by the Attending Veterinarian based on the health report. Imported animals will undergo quarantine for specific

period of 6 to 7 weeks. Once the animals are declared disease free, they can be moved into the holding rooms.

- **Use of animals**
 - All cages must have a completely filled cage cards including the protocol number that is current and contact information.
 - Strictly following specific guidelines like: appropriate use of procedure rooms, use of biosafety cabinets, recapping of needles, and disposal of animal carcass.
 - Under no circumstances, live animals can be taken to the laboratory areas. Only tissues free of skin can be taken to the labs. Moving animals from one room to another is also prohibited.
 - Timely weaning is required to avoid overcrowding. If notified of overcrowding, correct within 24 hours to avoid extra charge.
 - **Preplanning** of experiments is an essential component in animal studies.
 - Be notified that animal care staff will perform Post Approval Monitoring to ensure that procedures are carried out properly and the protocol is strictly followed.
 - Any special situations that require additional inputs should be brought to the attention of animal care staff.

B. Animal Facility Tour/Demonstration: (Includes hands on training and demonstration of some selected procedures)

Entry into Animal facility:

- Donning of PPE (Personal protective equipment): Lab coats, face masks, shoe covers, gloves and hair bonnets. Disposable lab coats can be labeled and saved for reuse. Facility staff will discard all used lab coats on a weekly basis, usually Monday morning. Disposable lab coats must be discarded if they are visibly contaminated or wet and soaked through.
- Restricted Access: No visitors are allowed into the facility. Request if needed. (Note visitor policy)
- Photography is not permitted. Contact the director of the facility for more information. (Note photography policy)

Animal Facility Basics

- **Five animal holding rooms, including BL2. In addition, one quarantine room.**
 - **Animals are housed in these rooms**
 - **Activities in these rooms include**
 - **Observation of animals**
 - **Setting up breeding**
 - **Weaning, identification, tissue sampling**

- **Minor procedures such as injections**
- **Three procedure rooms**
 - **These rooms must be reserved in advance; calendars are accessed through the intranet.**
 - **Procedure Room 1 – For gaseous anesthesia and for any surgical procedures. No bacteria allowed.**
 - **Procedure Room 2 – For euthanizing animals and for tissue necropsy**
 - **Procedure Room 3 – This room has a special surgical microscope and is mostly used for pulp exposure, ligature placement etc. Bacteria are allowed in this room. Before use, make sure to cover the handles, levers and knobs with suitable sleeves and tapes to ensure sterility. A sign in/out form is available for use by the staff.**
 - **BL2 room used for hazardous procedures and for procedures with bacteria requiring isofluorane**

Animal Holding Room Demonstration:

1. Cage Cards: All cages must have a completely filled cage card including the Protocol Number. Information must be written very clearly. Colored cage cards are used by animal facility staff for special situations:

Yellow – Overcrowding

Green – Sick or dead animals

Pink – Birth records

Red – Sentinel cages

2. SPF Conditions: All mice at Forsyth are housed under specific pathogen free conditions. This means that the facility is free of a specific set of viruses and bacteria that cause disease in mice such as mouse hepatitis virus. The presence of these diseases is a problem for many research procedures, especially immunological experiments. To maintain this status, mice are maintained under sterile conditions.

Mouse cages are only opened in the biosafety cabinet or change station

All Cages are autoclaved before use

Sterile food is provided

Enrichment items are sterilized before use.

These items used for mice should only be opened in the biosafety cabinet:

Regular Food

Breeder Food

Nestlets

Enrichment items

3. Bio Safety cabinets: All the holding rooms are equipped with certified bio safety cabinets/change stations. All manipulations of animals must take place inside the cabinet Staff should familiarize with the use of these units. Sash height, lights, UV lights, ventilation etc. Use of the biosafety cabinet and the concept of maintaining a sterile area during examination or cage change are very important. Use of Clidox/ethanol at all times – spray surface and hands. Cage lids upside down. Use sterile paper towels. Follow any instructions posted on each cabinet. Units must be cleaned with 70% ethanol after each use.

4. Housing:

- Mice – Standard 70 sq. in cage will carry a maximum of five adults.
- Rats – According to body weight.
- Large breeder cage – One male and two females.

Demonstration: Setting up a cage (bar lid, adding food in a sterile manner, adding nestlet and igloo in the cage change station, add card holder, fill in card. Use of Clidox.

5. Handling and Restraining of Animals:

Demonstration: Handling mice - Transfer mice from one cage to another (clean cage just set up). Demonstrate picking up the animal to check its ear tag or ear punch. Demonstrate sexing.

- Mouse is allowed to rest on a wire bar lid and handled by the base of the tail. Holding the tail with one hand and scuffing the skin in the neck region with the other hand, the animal can be restrained easily for closer observation, examination, sexing and for administering injections.
- Intravenous injections are carried out via tail vein and needs sedation and more experience. It will be demonstrated at a later time on a need basis.
- Small rats are restrained the same way as the mice. Adult rats are restrained using special restraint devices or with the use of gaseous anesthesia.
- Any injury, animal related or otherwise should be brought to the attention of animal care staff and work related incident report filed. If needed, medical attention is sought through appropriate channels.
- Hamsters, Guinea Pigs and Rabbits: With these three groups, animals are not available at all times. Any training related to these animals are done when needed and not during the routine training times.
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6. Breeding

- Standard cages can only have one male and one female.
- Special Breeder food has higher fat content – place sign on cage.
- Gestation is usually 19-20 days, depends on species. Animal facility staff will put a pink card giving date of birth.

- Weaning must be done between 23-28 days. After this the cage will be considered overcrowded.

Demonstrate sexing/weaning if possible.

7. Identification of Animals:

- Animals are identified using ear tags or by making ear punch with a special instrument applicator. Under light gaseous anesthesia, ear tagging or ear punching can be carried out with no stress to the animals. Correct placement of the ear tag is important and it comes with experience.
- Ear punching and tagging is demonstrated during specially arranged session.

8. Procedure Room 1:

- **Isofluorane apparatus: mixes appropriate amounts of anesthetic (isofluorane) and oxygen. Induction in chamber, nose cone for procedures. Isofluorane is hazardous to humans. Level of exposure controlled by charcoal canister. Critical to follow instructions completely.**

Administration of anesthesia is demonstrated during specially arranged session.

- Injectable Anesthesia – In most of the rodent species it is achieved by using a combination of ketamine and xylazine at the appropriate dose. It is calculated according to the body weight of the animal and species. These are controlled substances and the staff should request in advance from the Director of the facility. All personnel must follow safe injection practices and should be familiar with it. This will be demonstrated during special session.
- Biosafety cabinet – used for procedure requiring sterility.
- Dexa Scan – mention hazard, may not be inside the room when it is in use.
- Location of surgical and other supplies in cabinet.
- Location of surgical log books.
- **No bacteria allowed in this room.**
- No recapping of needles.
- Clean and disinfect (use Clidox) after use.

9. Surgical procedures:

All approved surgical procedures are described in the appropriate animal protocol. All staff must follow the Forsyth Aseptic Surgery Policy. The staff performing the surgery should be experienced and be very familiar with the steps and procedures and prepare in advance before the surgery. Make sure that all items needed are available including anesthesia, pain medication. These procedures are carried out in a dedicated room, Procedure Room 1 and all such surgical procedures are recorded in a surgery log book. In addition, close follow up observations of animals that underwent surgery are recorded for one week. Record the

administration of pain relieving medications. During surgery and post-surgery, it is advised to use a heating pad as the animals tend to lose their body temperature. A suitable heating pad with circulating warm water is available for use. Recovery period should be closely monitored before bringing the animals to the holding room cage.

10. Procedure Room 2: Euthanasia and Necropsy

- Small animals, mice and rats are euthanized using carbon dioxide gas. As much as possible, home cage should be placed inside the euthanasia chamber with the animals inside, then the gas valve opened for continuous flow. The flow of gas is gradual and the flow valve restricts to the set limit. This is to avoid any sudden surge of gas that can result in stressing the animals. The animals are closely observed for complete cessation of breathing. The gas flow should continue for more than a minute beyond apparent death.
- Euthanizing of animals will be demonstrated and each participant given the opportunity to perform one time.
- In adult animals, it is easier to confirm death and if needed, the thoracic cavity can be opened to confirm the euthanasia. For mice less than 4 weeks old, it is necessary to open the chest cavity or completely decapitate the head to confirm death after deep narcosis. This is demonstrated during training.
- Follow the instruction sheets posted near the euthanasia chamber. Euthanized animals are bagged and placed inside the dedicated freezer in the 4th floor.
- Any animals earmarked for euthanasia, can be clearly identified with a sign “Euthanize” and **left in the rack**. Animal care staff will take care of this. **Cages with animals should not be left outside the racks in the hallway with the sign “Euthanize”**.
- Room is also used for procedures in animals after death such as removal of tissues. Location of supplies (bench covers, plastic bags etc.)
- Clean up and disinfect (use Clidox) after use.

11. Procedure Room 3:

- Used for procedures inoculating bacteria in the oral cavity.
- SOP for surgical microscope
- Clean up and disinfect (use Clidox) after use.

12. Cage Wash room/Clean cage room:

- **What to do if you need supplies and there is no one around:** Items needed for cage establishment are available in all the animal rooms. During training, these items will be identified and the staff must familiarize with the needs.
- **Where to leave dirty cages:** Dirty cages must be left outside the cage wash area.

13. Safety Considerations:

- Contact information is available near the phone inside the animal facility.
- In case of injury, contact the director of animal care, the IACUC chair or the Safety Officer.
- If working alone after hours or on the weekend, set up a text-in/text-out system with your supervisor or senior colleague.

14. Beetle Room:

To de-flesh and to recover bone samples: Identify sample cups with name, sample no. and place the cups carefully and prevent from tipping over. Remove samples when complete.