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UC Davis
Institutional Animal Care and Use Committee (IACUC)

Title: *Rodent Surgery Guidelines*

I. Purpose:

This document describes the current requirements for performing rodent survival surgery.

II. Policy:

General requirements for rodent survival surgery include:

1. Surgical area selected documented in an approved Animal Care and Use (IACUC) protocol
2. Sterile instruments
3. Aseptic technique
4. Anesthesia and analgesics as described in an approved IACUC protocol
5. Monitoring plan to ensure animal wellbeing as documented in an approved IACUC protocol
6. Post-operative care
7. Records documenting all activities
8. Training of personnel documented prior to initiating surgery

Investigators may conduct rodent survival surgery in IACUC-approved investigator laboratories under special circumstances; however, use of an animal facility procedure room is preferred. While there is no requirement for a dedicated surgical facility for rodents, ***there are requirements for how rodent surgery must be conducted.***

Major survival surgery: penetrates and exposes a body cavity, produces substantial impairment of physical or physiologic functions, or involves extensive tissue dissection or transection (e.g., laparotomy, thoracotomy, joint replacement, limb amputation). Cranial implants and craniotomies that do not open the dura mater other than creating small holes for needles or small cannula shall be considered minor surgical procedures since the brain (e.g., cranial cavity) is not exposed and no substantial physical impairments are produced. Examples for minor cranial surgeries could include neurorecording chamber placement and infusion/injection cannulae placement. Any

cranial surgery that involves incising the dura mater shall be considered a major operative procedure.

Laparoscopic procedures that do not produce substantial impairment or involve the removal of organs or large sections of an organ such as oocyte collection, tissue biopsy, visual evaluation for sex determination shall be considered minor operative procedures. Conversely, laparoscopic procedures that include organ removal or partial organ removal such as ovariectomy, cholecystectomy, hepatic lobectomy shall be considered major operative procedures. The ultimate decision to classify a surgery as major or minor will be made by the IACUC. Contact the IACUC office for additional details regarding major versus minor surgical classifications.

III. Procedure:

A. Acclimation Period: Per the Guide for the Care and Use of Laboratory Animals (*The Guide*), newly received animals should be provided a period for physiologic, psychological, and nutritional stabilization and acclimation before their use.

Rodents should have a minimum 3-day acclimation period prior to anesthesia, surgery, or similarly invasive procedures.

It is the responsibility of the Principle Investigator (PI) to ensure the acclimation period has been met. Additional information can be found in the IACUC Policy "[Animal Acquisition, Acclimation and the Animal Tracking System](#)".

B. Preparation for Surgery: Prior to and between surgeries, clean and disinfect the surface upon which surgery will be performed. Use soap and water, rinse thoroughly, and follow with an appropriate disinfectant (e.g., peroxygen compounds, 10% diluted bleach, chlorhexidine, or quaternary ammonium compounds). Disinfectants must be prepared and used according to the manufacturer's recommendations to ensure appropriate dilution and contact time for disinfection.

C. Use of Sterile Instruments: Surgical instruments must be sterilized for use in survival rodent surgery. Several techniques (e.g., steam, dry heat, gas sterilization or chemical agents) can be used to sterilize instruments and other materials that will come in contact with animal tissues. **Steam or dry heat are the preferred methods to sterilize surgical instruments.** Another acceptable method of instrument use includes "tips only" practice. Using clean exam gloves and a "[tips-only](#)" technique restricts you to using only the sterile working ends of the surgical instruments to manipulate the surgical field.

Chemicals: Chemicals used to sterilize surgical instruments **must be classified as a sterilant not a disinfectant.** Chemical sterilants typically require a contact time of 6-24 hours, depending on the chemical used. For example, chlorine dioxide requires a

minimum of 6 hours of contact time. Glutaraldehyde and Cetylcid require instruments be soaked a minimum of 10 hours. All **instruments sterilized by chemicals must be rinsed in sterile water before use in tissues**. In between uses, containers used to store these surgical instruments during the sterilization time (see above) will be autoclaved (if metal) or gas-sterilized (if plastic-type material). Overall, chemical sterilants must be prepared and used according to the manufacturer's recommendations.

Multiple Surgeries: When performing surgeries on multiple animals it is recommended to have at least 2 sets of sterile instruments to allow re-sterilization of instruments between animals. Chemical sterilants typically require hours of contact time, therefore they are seldom practical for re-sterilizing instruments on the same day as surgery. It is suggested that a new sterile instrument pack be used after every 6 major surgical procedures.

Glass bead sterilizer: The optimal method for re-sterilization of instrument tips on the day of surgery is using a Glass bead sterilizer. While the first set of instruments is being re-sterilized, the second set is used. After using a set of instruments, remove all organic material and then immerse the instruments in a glass bead sterilizer for 20-30 seconds (follow manufacturers guidelines). **Make sure the tips of the instruments have cooled before use on animals.** Tips may be cooled by dipping in sterile water. ***It should be noted that glass bead sterilizers and tips of instruments sterilized in glass bead sterilizers are capable of producing severe burns. Care must be exercised when using a glass bead sterilizer, and all manufacturer instructions and safety precautions must be followed to avoid injury.*** With each use, the beads will be visually inspected to ensure there is no residual, contaminating material from previous use. Beads must be replaced as needed based on individual manufacturer recommendations.

D. Aseptic Technique:

Preparation of the Animals: While under anesthesia (as approved in an IACUC protocol) and prior to transporting the animals to the surgery area, it is suggested that at least a centimeter of hair be removed on either side of the incision site. Hair can be removed by clipping with an appropriate sized clipper, shaving with a razor, plucking (in anesthetized rodents), or by using a depilatory cream. Loose hair is then removed with a dry gauze or careful vacuuming with a HEPA filtered vacuum system. Depilatory cream must be thoroughly removed using warm water/saline. If the cream is left on the rodent's skin for too long, it can cause chemical burns and severe discomfort. Place lubricating ophthalmic ointment (such as Lacrilube® or Purilube®) in the anesthetized animal's eyes to prevent drying of the cornea.

Clean and aseptically prepare the surgical site: Use an effective antiseptic surgical scrub (e.g., Nolvasan®, Betadine®). Carefully scrub the area with a new clean surgical sponge or sterile cotton swab. Scrub in a gradually enlarging circular pattern from the center of the proposed incision to the periphery. The sponge or swab should not be brought back

from the contaminated periphery to the clean central area. Repeat with a 70% alcohol (or sterile water) soaked sponge or sterile cotton swab. **Repeat this process three times** to minimize the presence of micro-organisms.

Preparation of the Surgeon: Surgeons must wash their hands with a surgical scrub (e.g., Betadine Scrub®, Nolvasan Scrub®, Avagard™) **wear a mask, sterile gloves, and clean cuffed lab coat or long cuffed sleeve scrubs.** Long hair must be pulled back. A new pair of sterile surgical gloves must be used for each animal.

During Surgery: The surgical field must be kept sterile throughout the procedure. Sterile instruments must be prevented from contacting non-sterile surfaces. Instruments must be placed on a sterile surface when not in use. In most cases, the use of sterile drapes is required for maintenance of the sterile field.

Implantable materials: All implantable materials must be sterile prior to insertion. This includes, but is not limited to, implantable devices such as osmotic pumps, catheters, suture material, or internal clips.

- E. Monitoring and care of animal well-being:** Monitor the animal carefully during the surgical procedure. Anesthetized animals must not be left unattended during the procedure. The animal's depth of anesthesia must be assessed by toe/tail pinch prior to making an incision. Surgeons must pay close attention to the animal's respiratory rate, body temperature, and response to physical stimuli when anesthetized. Evaluating the animal's response to surgery (e.g., increased respiratory rate, movement, or vocalization) will also help determine the anesthetic depth.

Maintain Body Temperature: To prevent hypothermia, do not wet the animal any more than necessary. Care should be taken to prevent contamination of the sterile surgical field during subsequent handling and positioning of the animal. Place the animal on a clean absorbent surface and maintain body temperature using a circulating water blanket, warm water bottle, or equivalent external heat source, taking care to not cause thermal burns to the animal's skin. *Traditional heating pads are not acceptable due to the risk of thermal burns.*

- F. Post-operative Care:** Recovering animals should **not** be placed onto loose bedding material until they are fully awake, as suffocation can result. A clean surface (e.g., paper towel) may be placed between the bedding and the animal until it is fully awake. Prevent hypothermia by placing the recovering animals in a warm cage. If necessary, the cage may be placed on a padded surface and supplied with supplemental heat as required (such as a circulating hot water pad). It is recommended to only put a heat source below half of the cage. Be cautious with supplemental heat sources; hyperthermia can be as detrimental as hypothermia.

Observe Animal: Animals must be in an area where they can be frequently (every 10-15 minutes) observed until they are ambulatory and fully awake. To prevent cannibalism or suffocation, it is best to separate non-ambulatory from ambulatory rodents. Once animals return to standard housing, the cage should be identified with an index card reading "Post-Op". Post-Op cards can be obtained from the facility manager and husbandry staff. The incision site must be assessed daily to ensure it is clean, dry, and intact. If the animal's incision dehisces, the PI or PI's staff must contact Campus Veterinary Services (CVS) for assessment and surgical repair. Animals must be observed for signs of post-surgical pain (e.g., vocalization, persistent lethargy, lameness, or other signs identified in the IACUC protocol). They may be treated per the approved protocol or Campus Veterinary Services (CVS must be notified. Notify CVS if signs of pain do not resolve after approved post-op treatment.

Maintain Hydration: Dehydration can be ameliorated by the administration of appropriate fluid therapy. Initially this may be performed by administering 1 to 2 ml of warm fluids (0.9% NaCl or equivalent) per 100 grams of body weight by subcutaneous injection. If blood loss occurred during the surgical procedure, or if the animal is slow to recover from anesthesia, provide additional fluids. Consult CVS for assistance with fluid therapy.

Daily Post-op Checks: The PI, a member of the investigator's staff, or other individual to whom post-surgical care has been delegated must check post-surgical animals at least daily for a minimum of 7 days, until wounds have healed, and/or sutures/staples have been removed, unless the protocol has been approved for shorter duration for minor procedures. Analgesic drugs must be administered at a dose and frequency needed to maintain an appropriate analgesic efficacy as specified in the approved IACUC Protocol. **Multimodal analgesia is generally recommended, and deviations from this must be scientifically justified and approved by the IACUC.** Animals should be observed for signs of pain or discomfort. Please review the Grimace Scales for assessing pain in [mice](#) and [rats](#). They must be treated per the protocol or CVS must be notified. Notify CVS if signs of pain do not resolve after approved treatment.

Antibiotics: In general, intra- or post-operative antibiotics are unnecessary when aseptic technique is maintained. If inadvertent contamination occurs during surgery, consult with CVS immediately. If routine post-operative antibiotics will be needed, their use must be included in the approved Animal Care and Use Protocol.

G. Records: Post-surgical records must be kept in the room where the animals are housed for the duration of post-operative care. After post-op is complete records must be retained in the lab for three years past the expiration date of the protocol per "[IACUC Records Retention Policy](#)". An example of an anesthesia/surgery record and the policy for record documentation can be found on the CVS [website](#).

Documentation: All daily observations and treatments must be recorded on the animal's post-surgical record. External wound clips, staples, or sutures must be removed when surgical incisions are healed, 7-14 days after the surgery, or as outlined in the approved Animal Care and Use Protocol. Consult with CVS if you have questions regarding the optimal staple or suture removal time. CVS must be notified if post-surgical complications occur.

H. Training: PIs are required to ensure they and all staff conducting or assisting with rodent survival surgeries are appropriately trained and that training has been documented. Researchers conducting surgical procedures must have appropriate training to ensure that good surgical technique is practiced – that is, asepsis, gentle tissue handling, minimal dissection of tissue, appropriate use of instruments, effective hemostasis, and correct use of suture materials and patterns. Personnel conducting survival surgery **must** complete the online [UC Davis Rodent Survival Surgery](#) course. Personnel must obtain training on aseptic techniques from other knowledgeable personnel or by attending the [Aseptic Techniques](#) instructor led training. PIs and staff may also receive hands-on training for specific surgical techniques from others proficient in the procedure.

Training Opportunities:

1. An [Aseptic Technique course](#) and an online [UC Davis Rodent Survival Surgery](#) course are offered by the IACUC Office.
2. There is also an [AALAS Learning Library](#) online module for Aseptic Technique for Rodent Survival Surgery.

Please contact the IACUC office for questions regarding training or to obtain an AALAS Learning Library username/password (iacuc-staff@ucdavis.edu).

I. Non-Survival Rodent Surgeries: Non-survival surgery is defined as any surgical intervention in which the animal will not recover from anesthesia. AVMA approved methods of euthanasia are exceptions to this definition. Transcardial perfusion and exsanguination by cardiectomy are not considered non-survival surgeries. It may not be necessary to follow all the techniques outlined in the section for survival surgery if non-survival surgery is performed, however, at a minimum, the surgical site should be clipped, the surgeon should wear a lab coat and gloves, and the instruments and surrounding area should be clean. For non-survival procedures of extended duration, attention to aseptic technique may be more important to ensure stability of the model and a successful outcome.

J. Contact Campus Veterinary Services (CVS) (530-752-0514 or lahc@ucdavis.edu or CVSvets@ucdavis.edu) for questions regarding animal health, anesthetic support, surgical wound care, postoperative analgesia, or other questions regarding these guidelines.

IV. Resources:

1. ILAR, Guide for the Care and Use of Laboratory Animals
<http://nap.edu/12910>
2. Animal Welfare Act and Regulations
<https://www.nal.usda.gov/awic/animal-welfare-act>
3. IACUC-52 "Surgery Guidelines for Non-rodent USDA Covered Species"
<https://research.ucdavis.edu/wp-content/uploads/IACUC-52.pdf>
4. IACUC-40 "Animal Acquisition, Acclimation and the Animal Tracking System"
<https://research.ucdavis.edu/wp-content/uploads/IACUC-40.pdf>
5. "Editable Rodent Anesthesia and Post Op form"
https://research.ucdavis.edu/wp-content/uploads/2015-UCD-Rodent-Surgery_Anesthesia-Record.docx
6. "Aseptic Technique In-Person Course"
<https://research.ucdavis.edu/research-support/animal-care-use/training-classes/aseptic-technique/>
7. "UC Davis Rodent Survival Surgery"
<https://research.ucdavis.edu/research-support/animal-care-use/training-classes/uc-davis-rodent-survival-surgery/>
8. AALAS Learning Library
<http://www.aalaslearninglibrary.org/>
9. NC3R^s "Mouse Grimace Scale"
<https://www.nc3rs.org.uk/3rs-resources/grimace-scales/grimace-scale-mouse>
10. NC3R³ "Rat Grimace Scale"
<https://www.nc3rs.org.uk/3rs-resources/grimace-scales/grimace-scale-rat>
11. "Techniques in aseptic rodent surgery"
<https://pubmed.ncbi.nlm.nih.gov/18729061/>
12. "IACUC Records Retention Policy"
<https://research.ucdavis.edu/wp-content/uploads/IACUC-12.pdf>