

**Animal Studies Committee Policy**  
**Rodent Survival Surgery**

**ASC Policy:**

To optimize animal health and well-being, survival surgery in rodents must be performed using sterile instruments, surgical gloves, masks and aseptic procedures<sup>1</sup> to minimize microbial contamination of exposed tissues.

**Guidelines:**

These guidelines apply to all rodent survival surgical procedures performed at Washington University.

**General considerations for rodent surgery:**

**Training:**

1. Research personnel must be appropriately qualified and trained in all procedures to assure that good surgical technique is practiced.
2. Good technique includes asepsis, minimal dissection and gentle tissue handling, appropriate use of instruments, effective hemostasis, and proper wound closure.
3. DCM provides rodent surgery and aseptic training wet labs free of charge and also some videos of specific techniques.
4. **Participation in the wet lab training is now required for all new staff and students entering a lab where survival rodent surgery is part of the job description.**
5. Cadaver practice prior to using live animals is strongly recommended.

**Animal:**

1. Rats and mice have a high surface to body volume ratio and rapid metabolism. Therefore pharmacological doses are generally higher than in larger species.
2. Dehydration can occur much quicker than in larger species. Subcutaneous (SC) fluid administration (e.g. saline) is recommended for most procedures.
3. Rodents lose body heat very rapidly during surgery. Providing a supplemental heat source is necessary to help maintain normal body temperature.
4. Preoperative fasting is not necessary unless alimentary surgery is to be performed.

**Surgical location:**

1. A dedicated surgical facility is not required for rodents.

2. The area in which surgery is conducted should be free of clutter and disinfected before beginning the surgical session.
3. This area should be located away from air supply ducts or other drafts to minimize hypothermia of the animal and accumulation of dirt and dust contamination on surfaces.
4. Access to the area by personnel not directly involved in the surgery must be limited when surgery is being performed.

### **Aseptic Technique:**

According to the *Guide for the Care and Use of Laboratory Animals (Guide)*,<sup>1</sup> survival surgery on rodents should be performed using aseptic procedures. At Washington University, aseptic technique is required for all rodent survival surgeries. Aseptic technique is a set of specific practices and procedures performed under carefully controlled conditions with the goal of minimizing contamination by pathogens. This includes preparation of the animal, preparation of the surgeon, sterilization of instruments, supplies, implanted materials, and use of other operative techniques to reduce the likelihood of infection. All individuals performing surgery must be appropriately trained in aseptic techniques to ensure that good surgical technique is practiced.

**Note:** *The principles of aseptic technique mentioned above must be adhered to if the animal is to recover from anesthesia, regardless of whether the endpoint is 30 minutes or 30 days.*

### **Pre-Operative Preparation:**

#### Surgical instruments and supplies:

1. Instruments and supplies must be completely sterilized before surgery begins by an acceptable method (e.g. autoclaving, gas sterilization).
2. Use of a bead sterilizer as the sole means of sterilization is considered an exception and requires ASC approval. Bead sterilizers are generally reserved for sterilizing instruments between rodents undergoing surgery the same day.

#### Animal:

1. Anesthetize the animal as specified in the ASC protocol.
2. Apply sterile non-medicated ophthalmic ointment to eyes to prevent corneal drying.
3. Perioperative analgesics must be administered and specified in the protocol.
4. Perform preoperative preparations (e.g., hair removal) in a separate area from where the surgical procedure will be conducted. Avoid clipping excess hair as this may exacerbate hypothermia.
5. Prepare the surgical site(s) with an appropriate skin disinfectant, such as Betadine and/or Nolvasan. **Alcohol alone is not regarded as a sole agent for disinfection.**
6. Skin disinfectants should be applied in three alternating cycles, starting at the incision
  - a. Line in a circular motion and moving outward in concentric circles.
  - b. This is followed by a skin disinfectant solution.
  - c. Avoid excessive wetting of the animal as this may exacerbate hypothermia.

7. Place a sterile drape over the animal to prevent contamination of the surgical instruments/supplies and to assure a sterile field at the surgical site.

Surgeon/Assistant preparation:

1. At a minimum, surgeons must wear a face mask and wash/dry their hands thoroughly before donning sterile surgical gloves.
2. Surgeons should wear clean scrubs or a lab coat and a surgeons cap if they will be working over an open incision and/or if they have longer than shoulder length hair.
3. Assistants working in the immediate vicinity (but not in the sterile field) should wear clean scrubs or lab coat and a face mask.

**Intra-Operative Procedures:**

1. The animal(s) must be maintained in a surgical plane of anesthesia (identified by a depressed blink reflex when the eye is touched or lack of withdraw when the toes are pinched) throughout the procedure and vital signs (e.g. respiratory rate, heart rate, skin/mucous membrane color, etc) monitored as appropriate.
2. Anesthetic/pain assessment (toe pinch, respiratory rate/quality, etc) should be done before making an incision and routinely thereafter to assure an adequate plane of anesthesia.
3. A sterile drape or tray should be used to provide a sterile area in which to place instruments/supplies on during surgery.
4. Instruments sterilized initially by autoclave or gas sterilization may be used for a series of similar surgeries, provided they are either bead sterilized or disinfected using an intermediate to high level disinfectant between animals.
5. Contaminated gloves must be changed between animals. Alternatively, if precautions are taken to minimize contamination, gloves can be disinfected with an intermediate to high level disinfectant and thoroughly rinsed with sterile water before the next animal.
6. Fluids given parenterally or used as lavage during surgery must be sterile. Warming fluids prior to administration is recommended. Fluid administration may be repeated post-operatively to replenish blood or fluid loss.
7. Proper surgical technique must be practiced which includes, asepsis, gentle tissue handling, minimal tissue dissection, appropriate use of instruments, and effective hemostasis.
8. Close surgical wounds using appropriate techniques and materials. Closure of incisions in body cavities should be in at least two layers, the first including the body wall and the second, the skin.
  - a. Deep tissue layers should be closed using absorbable suture material.
  - b. For skin closure, use non-capillary suture such as nylon, polypropylene, polyethylene, stainless steel wire or metal wound clips.
  - c. The use of a continuous suture pattern in the skin layer is discouraged because one break in the suture line can result in wound dehiscence (wound splits open).
  - d. Rodent skin has a tendency to invert, therefore everting patterns such as a horizontal mattress or simple interrupted suture pattern should be used.

- e. Skin should not be closed with capillary (absorbable) suture (e.g. such as silk, Dexon or Vicryl) since rodents generally reside on contaminated surfaces and may subsequently develop wound infections.
- f. When applying tissue glue for skin closure it's important to apply only a very small amount using a smaller gauge needle (e.g. 25 gauge or smaller). Applying too much glue can irritate the skin and cause the animal to chew at the incision site.

### **Post-Operative:**

1. Move the animal(s) to a warm, dry area and monitor during recovery. Monitoring should include observing the animal's respiratory rate, skin/mucous membrane color, etc.
2. Animals must be monitored until they are able to move around or maintain a sternal position.
3. Provide fluids, analgesics, or other drugs as specified in the approved protocol or as directed by the veterinary staff.
4. At a minimum, all post-procedural animals must be observed once a day. Animals should be checked for signs of pain, abnormal behavior/appetite, or infection (e.g. redness, swelling, discharge).
5. Signs of pain and distress include: decreased food/water consumption, weight loss, dehydration, tented skin, rapid open mouth breathing, biting, aggression, hypo/hyperactivity, unkempt appearance (piloerection, rough/dull hair coat), hunched posture, head pressing, sunken eyes, etc.
6. Seek veterinary assistance in case of complications (infection, wound dehiscence, etc).
7. Skin sutures must be removed as soon as the wound is healed, usually 7-10 days post-operatively.
8. Maintain a surgical record on rodent post-surgical monitoring card. These cards may be obtained from DCM managers/supervisors and provide information on type of procedure, who performed the procedure, date and end time of procedure, analgesia provided, number of animals involved, protocol number, emergency contact name and phone number, date of suture/staple removal, and any other relevant notes. The card should be maintained on the cage until sutures/staples removed or procedure is otherwise completed. Completed cards are to be maintained by the investigator for a period of two years to document that post-surgical monitoring was properly conducted.
9. Unconscious animals must not be left unattended.

### **Note on Non-survival surgical procedures:**

In non-survival surgery, an animal is euthanized before recovery from anesthesia. It may not be necessary to follow all the techniques outlined above if the duration of the experiment is brief (<4 hours); however, at a minimum, the surgical site should be clipped, the surgeon should wear gloves, and the instruments and surrounding area should be clean. For non-survival procedures of extended duration (> 4 hours), attention to aseptic technique may be more important in order to ensure stability of the model and a successful outcome.

### **References:**

- 1.) The *Guide for the Care and Use of Laboratory Animals*: 8<sup>th</sup> Edition. (*Guide*). National Research Council. 2011
- 2.) Animal Welfare Act (AWA)
- 3.) Mouse and Rat Anesthesia cocktail formulations:  
[https://researchsecure.wustl.edu/Offices\\_Committees/DCM/InfoServices/Pages/Rodents.aspx](https://researchsecure.wustl.edu/Offices_Committees/DCM/InfoServices/Pages/Rodents.aspx)
- 4.) Division of Comparative Medicine training module titled, “Rodent Anesthesia and Surgery”.  
<http://learnatwork.wustl.edu>