

Recommendations for Aseptic Technique and Post-Operative Care for Rodent Surgery

It is the responsibility of the veterinary staff, investigator, laboratory animal technicians, and the facility manager to ensure that all personnel performing the procedures outlined in this document are properly trained in the correct technique and that anesthesia, post-operative pain medication and care are provided to the animals. Where possible, surgeries should be performed during normal business hours and always evaluated the next day for post-operative complications. For training purposes, surgeries must be performed during normal business hours and evaluated the next day for post-operative complications. The following procedures are recommended to ensure adequate aseptic technique and post-operative care:

A. Animal Preparation

1. The hair over the surgical site should be clipped ~150% of the anticipated surgical area using #40 or #50 clipper blade, taking care not to cut the skin. This should be performed in an area separate from where the surgery is to be conducted.
2. Rodents should be anesthetized according to the Animal Study Proposal.
3. Once response to a gentle toe pinch is lost, anesthetic depth is sufficient for surgery; the animal's ears and feet, and mucous membranes of the eyes and nose should be pink indicating adequate oxygenation. Respiratory rate should be monitored throughout the procedure to ensure appropriate depth of anesthesia.
4. Artificial tears ointment should be applied for protection and lubrication if the animal is anesthetized for more than 5 minutes.
5. The surgical site should be scrubbed three with Betadine, Chlorhexidine, or another approved antiseptic scrub, alternating with 70% ethyl or isopropyl alcohol, which must be allowed to dry before incision. Sterile gauze must be used and scrubbing motion should begin at the anticipated incision site and spiral outwards [do not go back to the incision site with the same gauze]. Application of a betadine or chlorhexidine solution (not scrub) is recommended after the surgical prep scrub. Please see Table 1.
 - A. Avoid excessive wetting of non-surgical areas of the animal with alcohol or antiseptic as this can exacerbate hypothermia
6. The rodent should be carefully placed onto a warm surface (use of electronic heating pads is prohibited) and positioned for surgery.

B. Surgeon Preparation

1. Surgery should be conducted in a disinfected, uncluttered area that promotes asepsis during surgery. Please see Table 2.
2. Scrubs and personal protective equipment [as dictated by the facility requirements and including a mask] should be donned by the surgeon.

3. Hands should be scrubbed thoroughly with antibacterial soap and new clean gloves [disposable or sterile] should be worn.

C. **Surgical Instruments**

1. Start each surgery or series of surgeries with a new autoclaved pack of instruments
2. Between animals, the instruments should be cleaned of particulate matter and placed in a scientifically acceptable disinfectant solution or a glass bead instrument sterilizer. The instruments should be wiped dry prior to use. If a hot bead sterilizer is used, allow adequate time for the instruments to cool before use.
 - A. It is recommended that a maximum of 5 surgical procedures in series be performed with a single autoclaved pack of instruments. Please see Table 3.
3. After all surgeries are completed, the instruments should be thoroughly cleaned prior to packing for the autoclave.

D. **Surgical Procedure**

1. The animal must be maintained in a surgical plane of anesthesia throughout the procedure [i.e., absence of toe pinch reflex].
2. Surgical drapes are recommended where practical. The use of an adhesive, clear drape is particularly useful for surgery on small animals
3. Begin surgery with sterile instruments and handle instruments aseptically.
 - A. Unwrap sterile instruments making sure to only touch the outer surface of the wrap before washing hands or performing surgical scrub
4. When using “tips only” technique, the sterility of the instrument tips MUST be maintained throughout the procedure.
5. Instruments and gloves may be used for a series of similar surgeries provided they are maintained clean and uncontaminated between animals. Please see Table 4.
6. Monitor and/or maintain the animal’s vital signs as necessary
7. Absorbable suture material or electrocautery should be used to control bleeding.
8. When the ventral abdominal cavity is opened, the abdominal lining, [peritoneum], and muscle layer must be closed with an appropriate number [for the length of the wound] of absorbable sutures. The skin should be closed separately.
9. When the peritoneal cavity is opened from a dorsal approach [incision on the back], it is recommended that absorbable sutures be used to close the peritoneum prior to skin closure.
10. Please see Table 5 for wound closure selections.

E. **Analgesia and Postoperative Care**

- A. Provide analgesics as appropriate and as approved in your Animal Study Proposal. Refer to the *Guidelines for Perioperative Analgesia in Rodents*.
 - Local analgesia with 1-3 drops [approximately one drop for each centimeter of the incision size] of 0.25% bupivacaine placed into the incision prior to closure
- B. Systemic analgesia with injectable agents such as buprenorphine, [0.05-0.1

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mg/kg SC every 8 hours]

2. Analgesia MUST be administered for major surgery.
3. Analgesia should be outlined in the Animal Study Proposal. Please refer to the *ACUC Guidelines for Perioperative Analgesia in Rodents*. Some examples include:
 - A. Contact LAM for additional information on analgesics and dosages.
4. When surgery is completed the rodent should be moved to a warm draft free location. A heat source should be used to keep the animal warm. The animal should be monitored for excessive body temperature during this period. Care must be taken when using a heat lamp as they can produce extremely high temperatures and ocular damage.
5. The animal should be observed until it is awake enough to maintain itself in a sternal/upright position and then it should be returned to a clean cage.
6. All post-surgical animals should be observed at least daily for five days following surgery for any signs of illness or infection of the incision.
7. Autoclips should be removed within 14 days depending on how quickly the incision heals. Absorbable suture material is not removed.
8. LAM should be consulted on any animal that does not seem to be recovering well, exhibits signs of pain or distress, or develops redness, swelling, heat, or discharge at the incision site.

TABLE 1 – Skin Disinfectants

Agent	Examples *	Comments
Iodophors	Betadine®, Prepodyne®, Wescodyne®	Reduced activity in presence of organic matter. Wide range of microbicidal action. Works best in pH 6-7.
Chlorhexidine	Nolvasan®, Hibiclens®	Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses. Excellent for use on skin.

** The use of common brand names as examples does not indicate a product endorsement*

TABLE 2 – Recommended Hard Surface Disinfectants

Always follow manufacturer’s instructions for dilution and expiration periods.

Agent	Examples *	Comments
Quaternary Ammonium	Roccal®, Quatricide®	Rapidly inactivated by organic matter. Compounds may support growth of gram negative bacteria.
Chlorine	Sodium hypochlorite [Clorox® 10% solution] Chlorine dioxide [Clidox®, Alcide®, MB-10®]	Corrosive. Presence of organic matter reduces activity. Chlorine dioxide must be fresh; kills vegetative organisms within three minutes of contact.
Glutaraldehydes	Cidex®, Cetylcide®, Cide Wipes®	Rapidly disinfects surfaces.
Phenolics	Lysol®, TBQ®	Less affected by organic material than other disinfectants.
Chlorhexidine	Nolvasan®, Hibiclens®	Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses.
Hydrogen Peroxide [Clorox Healthcare®]	Clorox Healthcare® Hydrogen Peroxide Cleaner Disinfectant	Rapidly disinfects surfaces without harsh chemical fumes or odors.

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TABLE 3 – Recommended Instrument Sterilants

Always follow manufacturer’s instructions for dilution, exposure times, and expiration periods.

Agent	Examples *	Comments
Steam Sterilization [moist heat]	Autoclave	Effectiveness dependent upon temperature, pressure and time [i.e., 121°C for 15 minutes vs. 131°C for 3 minutes]
Dry Heat	Hot Bead Sterilizer Dry Chamber	Fast. Instruments must be cooled before contacting tissue. Only tips of instruments are sterilized with hot beads. Instruments require approximately 20 seconds of contact with the beads at a temperature of 240°C-270°C.
Gas Sterilization	Ethylene Oxide	Requires 30% or greater relative humidity for effectiveness against spores. Gas is irritating to tissue and requires specialized equipment for use. All materials require safe airing time.
Chlorine	Chlorine Dioxide	Corrosive to instruments. Instruments must be rinsed with sterile saline or sterile water before use.
Glutaraldehydes	Cidex®, Cetylcide®, Metricide®	Several hours required for sterilization. Corrosive and irritating. Instruments must be rinsed with sterile saline or sterile water before use.
Hydrogen Peroxide-Acetic Acid	Actril®, Spor-Klenz®	Several hours required for sterilization. Corrosive and irritating. Instruments must be rinsed with sterile saline or sterile water before use.

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TABLE 5 – Wound Closure Selection

Material *	Characteristics and Frequent Uses
Polyglactin 910 [Vicryl®] Polyglycolic Acid [Dexon®]	Absorbable. 60-90 days. Ligate or suture tissues where an absorbable suture is desirable.
Polydioxanon [PDS®] Polyglyconate [Maxon®]	Absorbable. Six months. Ligate or suture tissues especially where an absorbable suture and extended wound support is desirable.
Polypropylene [Prolene®]	Nonabsorbable. Inert.
Nylon [Ethilon®]	Nonabsorbable. Inert. General closure.
Silk	Nonabsorbable. Excellent handling. Preferred for cardiovascular procedures. Caution: Tissue reactive and may wick microorganisms into the wound.
Chromic Gut	Absorbable. Versatile material.
Stainless Steel Wound Clips or Staples	Nonabsorbable. Requires instrument for removal.
Cyanoacrylate [Vetbond®, Nexaband®]	Skin glue. For non-tension bearing wounds. The glue requires adequate moisture and pressure to properly bond wound. Please note that if too much glue is applied, an exothermic [burn] reaction can occur.

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Questions concerning this guideline should be addressed to the veterinary staff, facility management, or technical staff

Required training for aseptic surgery:

<http://www.jove.com/video/2586/principles-of-rodent-surgery-for-the-new-surgeon> (video)
<http://www.ncbi.nlm.nih.gov/pmc/articles/PMC2587003/> (article)