

**Arizona State University
Institutional Animal Care and Use Committee
STANDARD INSTITUTIONAL GUIDELINE**

RODENT SURGERY

It is the policy of the IACUC that all survival surgeries involving rodents be conducted using appropriate aseptic techniques as described in this guideline.

A. Aseptic Technique

1. General Information

- a. Although mice and rats have been touted as being resistant to post-surgical infections, existing literature documents how certain subclinical infections can influence study results or become clinical diseases following stress or immune suppression (Cooper et al, 2000).
- b. Rodent survival surgery must be performed using aseptic technique; which includes the preparation of the surgical surface, the animal, the surgeon, and the surgical instruments. The primary objective of this is to reduce microbial contamination of the incision and exposed tissues to the lowest possible practical level.
- c. Most rodent surgeries utilize aseptic tip technique. The tip of the instrument is the only sterile part of the instrument and the only item that should come into contact with the surgical site. The surgeon's fingers, even if gloved, should never touch the area of the animal that has been disinfected, internal tissues, the sterile tips of the instruments, the sterile drape, or any sterilized consumables.

2. Preparation of the Surgical Surface

- a. Before beginning rodent surgery, the laboratory bench or table where the surgery will be performed should be cleaned and disinfected. Quaternary ammonium compounds (e.g., Virex) are good choices for such disinfection.
- b. Locations where air currents and dust are difficult to control, such as in front of windows or next to doors, should be avoided as rodent surgery tables. Likewise, rodent surgery should not be performed in or in front of an exhaust hood unless it is required for biosafety.

3. Preparation of the Animal

- a. Enough fur should be shaved from the incision site to prevent wound contamination; the edges of the incision should not come in contact with fur. Due to the fur's ability to assist in thermoregulation, care should be taken not to remove excessive amounts of fur.
- b. The incision site should be disinfected first with either a dilute iodine solution (e.g., Betadine) or dilute chlorhexidine (e.g., Nolvasan), followed by 70% alcohol or sterile saline. This process should be repeated two additional times.

- c. Each step in the disinfectant-alcohol process should be applied in a concentric fashion beginning at the incision site and moving outward, away from the incision site to the periphery of the surgical field.
4. Preparation of the Surgeon
 - a. All individuals participating in surgery should wear a face mask and secure long hair.
 - b. It is recommended that all individuals performing surgery wash their hands before donning exam gloves. Sterile gloves are not needed during aseptic tip surgery since the surgeon's hands never touch sterile materials or internal body tissue. If the surgeon needs to disinfect their gloves between animals, exam gloves can be wiped down with 70% alcohol (Keen et al., 2010; LeMoine et al., 2015). If the surgeon's hands need to come into direct contact with the surgical site, then sterile surgical gloves are necessary.
5. Preparation and Use of Surgical Instruments
 - a. Surgical instruments for rodent surgery should be sterilized, ideally utilizing steam or gas sterilization. Hot bead sterilization is more practical, and equally effective, when only instrument tips will contact the animal. While inferior because it is not a sterilant, 70% isopropyl alcohol can be effective at preventing bacterial contamination (Huerkamp, 2002; Keen et al., 2010), and may be approved by the IACUC in some situations (e.g., field surgery on multiple animals without a source of electricity).
 - b. Rodent surgeries may be done several in succession. In such cases, it is imperative that instruments are cleaned and sterilized in a hot bead sterilizer between surgeries. Wipe any blood or tissue off the instruments before placing in the sterilizer. After sterilization, instruments should be cooled to room temperature prior to contacting the animal by soaking the sterilized instruments in sterile water or saline. Using a chemical disinfectant (e.g., quaternary ammonium compounds, iodine solutions, alcohol, and phenol compounds) instead of a hot bead sterilizer must be pre-approved by the IACUC.
 - c. Once sterilized, parts of the instruments that will touch internal tissue must only contact sterile surfaces. During surgery, but when not in use, instrument tips should be kept on a sterile surface or suspended so that they do not touch any surface. When using hot bead sterilization, the handles of the instruments are not being sterilized, so instrument tips should not come in contact with the handle of another instrument or touch a place where an instrument handle has previously been. If the tip of an instrument touches a non-sterile surface, it should be re-sterilized. If the instrument handle is placed on a sterile surface, that surface is no longer considered sterile.
 - d. It is recommended to use one pair of scissors and forceps for skin incisions and a second pair of scissors and forceps for abdominal wall incisions and manipulation of viscera. Alternatively, the instruments can be disinfected in a hot bead sterilizer after making the skin incision.
 - e. To avoid contamination of sterile items, it is recommended that the animal's body, other than the site of the incision, be covered with a sterile drape or sterile gauze. Glad Press'n

Seal can be used to cover the body to avoid gross contamination of the surgical site with hair or dander, but, as the wrap is not sterile, contact of it with sterile instruments must be avoided. Once applied to the animal, the Glad Press'n Seal can be disinfected around the incision site with either a dilute iodine solution (e.g., Betadine) or dilute chlorhexidine (e.g., Nolvasan), followed by 70% alcohol or sterile saline.

- f. If internal tissue needs to be exteriorized through the incision and will re-enter the body cavity, it must be placed on a moist sterile surface (e.g., a piece of sterile gauze dampened with sterile saline).

B. Facility

According to the *Guide for the Care and Use of Laboratory Animals*, a separate facility specifically designated for rodent surgery is not required. The *Guide* states that, "for most rodent surgery, a facility may be small and simple, such as a dedicated space in a laboratory appropriately managed to minimize contamination from other activities in the room during surgery."

C. Choice of Surgical Materials

1. Expired drugs cannot be used for any surgery. Expired supplies cannot be used on animals for any survival surgery.

2. Surgical Instruments

The size of the rodent must be considered when planning a surgical procedure. The surgical instruments (scalpel blade, scissors, forceps, etc.) need to be matched to the size of the rodent and the procedure in order to minimize trauma. The use of proper instruments and surgical technique are likely as important as the use of aseptic technique in preventing post-surgical infections (Cooper et al., 2000).

3. Suture Materials and Suture Patterns

- a. The smallest size suture appropriate for the tissue and rodent should be used (e.g., 3-0 or 4-0 for a rat, 4-0 or 5-0 for a mouse).
- b. Surgical Needles
 - (1) Cutting needles should be used when it is necessary to suture dense, difficult to penetrate tissue such as skin. Using cutting needles on thinner tissue (e.g., intestine) will readily tear the tissue during the placement of the suture.
 - (2) Non-cutting needles (e.g., taper point, round needles) are used primarily for suturing easily penetrated tissues such as peritoneum or intestine.
 - (3) Needle curvature options depends on the tissue being sutured. 3/8 circle is the most common shape utilized and is appropriate for most surgical applications. 1/2 circle is used in confined areas deep inside the body when organs cannot be easily exteriorized.

Reviewed 7/27/2023

Updated 7/23/2020

Updated 8/27/2015

- (4) Needle length is from the tip of the needle to the suture attachment site. Using the smallest needle length for the procedure allows for better handling and less tissue trauma (e.g., 19-24mm for a rat, 13-19mm for a mouse).
- c. Absorbable Sutures
- (1) Absorbable suture materials should be used on internal tissue when the animal is expected to survive long-term. It can also be used for small skin incisions in areas with limited potential for dehiscence, such as the back. Absorbable suture is preferred for field studies to avoid the need to recapture the animal or permanently leave sutures in.
 - (2) Examples of absorbable suture material appropriate for rodent surgery include polyglactin 910 (e.g., Vicryl), poliglecaprone 25 (e.g., Monocryl), and polydioxanone (e.g., PDS).
- d. Non-absorbable Sutures
- (1) Non-absorbable suture materials should be used whenever tension exists on an incision and the potential for dehiscence exists. Non-absorbable suture materials are appropriate for skin closure provided the suture can be removed in approximately 7-14 days.
 - (2) Examples of non-absorbable suture material appropriate for rodent surgery include nylon (e.g., Ethilon) and polypropylene (e.g., Prolene).
- e. Monofilament Sutures
- (1) Monofilament sutures are made of a single strand of material, allowing the suture to pass through tissue with less resistance. The single strand construction also reduces the amount of bacteria that might wick through the surgery site. This characteristic makes monofilaments ideal for skin sutures. Monofilaments have greater memory, making them harder to work with.
- f. Multifilament Sutures
- (1) Multifilament sutures are made from multiple strands of material twisted or braided together. This provides greater strength and flexibility. The downside to multiple strands is increased drag causing more damage to the tissue, increased surface area to harbor bacteria, and wicking potential to bring microorganisms into the incision.
- g. Suture Patterns
- (1) To avoid detrimental outcomes (e.g., internal hemorrhage, skin dehiscence), suturing skill must be well established prior to performing survival surgeries. Improperly tied knots can unravel, so care must be used when placing knots. After each throw, both sides of the suture strand should be pulled in opposite directions with even pressure to lay the knot flat. Each knot should have 3 throws.

- (2) The abdominal wall, and other high-tension critical tissues should be closed using a simple interrupted pattern rather than a single continuous running suture. This reduces the probability that a large defect will occur should a knot or tissue placement fail. The same is true for skin closure, but to a lesser extent since the skin has less tension, is less vulnerable to tearing, and can be visually inspected to detect problems early.
- (3) Rodents can chew sutures out, so it is advisable to use staples when closing the skin in areas the animal can reach with its mouth.

4. Staples and Surgical Adhesive

a. Surgical Staples

- (1) Surgical staples are an effective and time-efficient alternative to suture material for skin closure.
- (2) Surgical staples should be removed 10-14 days post-surgery, as their extended presence can cause irritation. To avoid skin damage, staples are best removed using a purposefully designed staple remover.

b. Surgical Adhesive

- (1) Surgical adhesive can be used for small skin incision (0.5 cm or smaller) or as an adjunct to close any gaps at the end of the incision previously closed with staples. When using to close the skin incision, bring the skin edges together until touching, then drop a small amount of the adhesive on top of the closed incision. Do not let the incision open when dropping the adhesive. Surgical adhesive should never be placed inside the incision. Only apply the adhesive to the incision site and not anywhere else on the animal.

D. Anesthesia

Consult the [Anesthesia SIG](#) for acceptable choices of rodent anesthetic agents.

E. Post-Surgical Care

Consult the [Post-operative Care SIG](#) for appropriate post-operative monitoring following rodent surgery.

F. Analgesia

Consult the [Analgesia SIG](#) for acceptable choices of rodent analgesic agents.

G. References:

Cooper, D. M., McIver, R., and Bianco, R. 2000. The thin blue line: A review and discussion of aseptic technique and post-procedural infections in rodents. *Contemporary Topics in Laboratory Animal Science* 39: 27-32.

Reviewed 7/27/2023

Updated 7/23/2020

Updated 8/27/2015

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- Keen, J. N., Austin, M., Huang, L. S., Messing, S., and Wyatt, J. D. 2010. Efficacy of soaking in 70% isopropyl alcohol on aerobic bacterial decontamination of surgical instruments and gloves for serial mouse laparotomies. *Journal of the American Association for Laboratory Animal Science* 49: 832-837.
- LeMoine, D. M., Bergdall, V. K., and Freed, C. 2015. Performance analysis of exam gloves used for aseptic rodent surgery. *Journal of the American Association for Laboratory Animal Science* 54: 311-316.