Institutional Animal Care and Use Committee (IACUC)

Policy on Surgery and Post-Surgical Monitoring of Rodents Used in Research

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Policy Statement

This policy applies to all surgical procedures performed on rodents at Indiana University in Bloomington (IUB) including post-surgical care and monitoring of animals for recovery procedures. Prior to performing any surgery techniques on laboratory animals, an approved IACUC protocol must be in place with appropriately trained personnel and procedures available. Specific procedures that can be used to remain in compliance with this policy can be obtained from your veterinarian or by reviewing related documents on the BIACUC website.

Reason for Policy

Survival surgery and post-surgical care of research animals are addressed in the Guide, PHS Policy and USDA Regulations. These documents specifically require that the Institutional Animal Care and Use Committee (IACUC) review surgical activities and the Attending Veterinarian (AV) oversee surgical

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procedures and post-operative care programs. All personnel involved with performing surgeries must be trained in basic surgical (aseptic) technique by an IUB veterinarian prior to performing surgery.

Definitions

**Surgery**: The incising of skin or mucous membrane with a sharp instrument for the purpose of treatment, removal of selected tissue, or implantation of experimental hardware e.g. implantation of a device in the body, removal of a tumor or other tissue, etc.

**Non-surgical Procedures**: Injections (IM, IV, IP, SC), urethral catheterization, MRI or ECG, external IV catheter placement.

**Aseptic Surgical Procedures**: Surgery performed using procedures that limit microbial contamination so that significant infection or suppuration does not occur.

**Major Survival Surgery**: Any surgical intervention that penetrates and exposes a body cavity (e.g. joint replacement, limb amputation, laparotomy, thoracotomy) or has the potential for producing a substantial or permanent physical or physiological impairment or involves extensive tissue dissection or transection in an animal that is expected to recover. The AV will determine whether a procedure is to be considered "major" or "minor" or simply a "procedure" in cases where it is not obvious.

**Minor Surgery**: Any surgical intervention that neither penetrates or exposes a body cavity nor produces permanent impairment of physical or physiologic function. Examples are superficial vascular cut down, and percutaneous biopsy. Minor surgeries may be performed under less stringent conditions than major procedures. For minor surgical procedures appropriate aseptic technique and postsurgical monitoring is still required.

**Multiple Major Survival Surgery (MMSS)**- These procedures on one animal are not allowed unless the procedures are related to the same project, justified in writing by the principal investigator for scientific reasons, and approved by the IACUC. Cost alone is not an adequate reason for performing MMSS procedures on an animal; however, such procedures may be justified in the interest of conserving numbers of rare species or as necessary to protect the health or well-being of the animal, as determined by the veterinarian.

**Non-survival Surgery**: surgical procedure where the animal is euthanized before recovery from anesthesia. At a minimum, the surgical site should be clipped, the surgeon should wear gloves, and the instruments and surrounding area should be clean.

**Sterilization**: The process whereby all viable microorganisms are eliminated or destroyed. The criterion of sterilization is the failure of organisms to grow if a growth supporting medium is supplied.

**Disinfection**: The chemical or physical process that involves the destruction of pathogenic organisms. All disinfectants are effective against vegetative forms of organisms, but not necessarily spores.

**Postoperative Monitoring**: Monitor an animal’s status by assessing physiologic parameters like heart rate, respiration, mucous membrane color, temperature, and incision site during the immediate postoperative period until the animal recovers from anesthesia and is able to return to the cage. Monitoring and provision of supportive care and analgesics for evidence of pain or distress should occur for a minimum of 7-10 days after the conduct of the surgery to ensure food/water intake, fecal/urine output, attitude, incision site healing, and ability to move in cage.

Regulatory Requirements for Surgery

1. All surgical procedures on animals must be approved by the Institutional Animal Care and Use Committee (IACUC).
2. **Rodent survival surgery** must be performed in a designated area using aseptic technique. This is a space in a laboratory or procedural room which is disinfected before use and during its use is designated only for surgery.

3. All individuals performing surgery must be appropriately trained to ensure that proper surgical technique is practiced including asepsis, gentle tissue handling, minimal dissection of tissue, appropriate use of instruments, effective hemostasis, and correct use of suture materials and patterns to prevent wound infection and promote wound healing.

4. Rodents do not need to be fasted before surgery, however if they are, it should be for short periods (4-6 hrs). The animal must be properly anesthetized for the entire surgery being unresponsive to surgical procedural stimulation and no withdrawal reflexes when rear toes are pinched.

5. The surgical field must be aseptically prepared and maintained for the performance of the surgical procedure. Appropriate preparation of the incision site and precise surgical techniques are required to reduce microbial contamination of the surgical site as well as the instruments and materials used.

6. Surgeons working with rodents should wear special surgical garb which at a minimum should include a surgical mask, surgical scrub top or clean gown or lab coat, sterile surgical gloves and hair cover. Safety glasses are highly recommended.

7. Surgical procedures using surgical instruments and implanted materials/devices that penetrate a body cavity or are placed beneath the skin of an animal must be properly sterilized prior to surgery. Specific sterilization methods should be selected on the basis of the physical characteristics of the materials to be sterilized and sterilization indicators should be used to validate that materials have been properly sterilized. Packs appropriately sterilized by the lab expire at 6 months after sterilization or sooner if the pack is partially opened or pouch is wet or opened.

8. It is the primary responsibility of the Principal Investigator to ensure that appropriate post-anesthetic/post-surgical monitoring is performed and that animal records are maintained as described below. It is the responsibility of the Principal Investigator or designate to monitor all animals during recovery from anesthesia.

9. If any animal develops unexpected surgical or post-surgical complications including death, the LAR veterinary staff should be notified immediately. Animals that die unexpectedly during or after surgery or are euthanized because of post-surgical complications must be preserved by refrigeration and must be available to the LAR veterinary staff for necropsy as soon as possible. Postmortem examinations will be performed at the discretion of veterinary staff. The PI will be notified of the results of the necropsy, including findings that indicate problems with the surgical technique, anesthesia/analgesia administration, or general health of the animal.

10. Animals who have been sedated with short-acting agents to induce chemical restraint (for example, use of ketamine/xylazine to chemically restrain rodents) should be monitored by the PI or designated lab staff until they become ambulatory. Only when ambulatory should they be returned to their primary housing location.

11. The PI/surgeon is responsible for maintaining accurate records regarding surgical procedures and perioperative care records. Group or individual records that detail procedures, drugs administered, dates, personnel, and pre- and post-surgical condition of the animal, and identification of the surgeon must be kept for groups of animals having undergone surgery. For individual rodents, individual records that provide the same details may be used in place of group records if procedures are complex or individual monitoring is needed. Regardless of species, monitoring (at least every 5-10 min.) is required from anesthesia induction until
recovery from anesthesia has occurred. Daily recorded observations are required until the post-operative period ends, the animal is eating and drinking normally, surgical wounds are healed, and the animal is acting normal for the species (usually 7-10 days after surgery when sutures are removed and surgical wounds are healed).

12. For the pre-, intra- and post-operative period, the investigative group should document the following (use LAR surgical/postsurgical records for this information):
   - Pre-anesthetic information (approximate weight of animals, pre-anesthetic drugs, etc.)
   - Brief description of the procedure (surgical or otherwise)
   - Time of anesthetic induction and intubation time (if applicable)
   - Anesthetic used, including dose (mg/kg), volume (ml), and route of administration (SC, IP, IV, etc.)
   - Duration of surgical anesthesia
   - Additional drugs and solutions administered
   - **Daily postoperative monitoring for 7-10 days** with the sutures or staples removed at the end of the observation period.

13. Records of active cases must be readily available to the personnel involved in postsurgical monitoring, the veterinary staff, the IACUC, and federal regulatory officials. Surgical records should be maintained near where the animal is housed. Only records of live animals present in housing areas must be readily available. **Surgical records are to be maintained by the lab for 3 years following the death of the animal or following the expiration of the related protocol.**

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**Veterinary and IACUC Responsibilities**

1. **Veterinarian’s Responsibilities:**

   The veterinarian shall provide the IACUC with assessments of the following:

   a. Preparation of the animal for the surgical intervention, to include the use of preanesthetic drugs where indicated, and appropriate anesthetic or analgesic agents;
   
   b. Appropriate preparation of the animal and use of aseptic procedures for the surgery;
   
   c. Adequate post-operative care, to include post-operative analgesics and supportive care where indicated is provided and;
   
   d. Veterinarian has implicit responsibilities to assess the potential for pain and distress that might be associated with the proposed animal activities, and to recommend the use of pain alleviating drugs, whenever possible, to alleviate those conditions.
   
   e. Veterinary staff involvement will be determined by the needs of the research project. Consideration will be made for the experience of the surgeon and research staff, the surgical procedure being performed, the species involved, and the needs of the convalescent animal. LAR veterinarians conduct initial veterinary evaluation of proposed surgical procedures and perioperative animal care when they participate in IACUC review of animal care and use protocols.
   
   f. Minimally, ongoing veterinary monitoring will consist of regular review of the research group’s documentation of perioperative care and observation of animals to ensure adequate postsurgical care. This will include review of surgery records, animal health records, and other documents relating to animal surgery. The LAR veterinary staff is available for
consultation when planning for postoperative care of animals and for postsurgical emergencies.

2. **IACUC Responsibilities:**

The IACUC evaluates proposed surgical procedures and perioperative care during the review of animal care and use protocols. Ongoing monitoring and oversight are exercised through periodic review of the animal care and use program, inspection of animal housing facilities and animal use areas, and reports from the veterinary staff and/or IACUC Manager. Ongoing projects that are found to be out of compliance with either this policy or the relevant protocol are subject to additional oversight, which may include suspension by the IACUC.

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**Materials**

**A. Instruments:**

1. Many routine supplies required for aseptic surgery can be purchased prepackaged and sterile (surgeon’s gloves, paper drapes, scalpel blades and suture material).

2. **All surgical instruments required for the procedure must be assembled, wrapped in packs or pouches and sterilized by autoclave, hydrogen peroxide, ethylene oxide or cold sterilant in accordance with the manufacturer’s recommendations prior to surgery.** If surgical instruments are going to be sterilized using an autoclave, the use of autoclave tape alone on the outside of instruments packs to monitor the effectiveness of such sterilization procedure is inadequate. Surgical packs must contain a sterilization indicator **INSIDE** of the pack. The outside of the pack should also be wrapped with autoclave indicator tape or have some type of external sterilization indicator (e.g., if using self-sealing sterilization pouches). The sterilization date and the initials of the person preparing the pack needs to be written on the outside of each pack. Instruments sterilized by labs have a **shelf-life of 6 months**. After such time the items must be re-sterilized in new packing material. If surgical packs or pouches have been opened, punctured or become wet, they are no longer considered sterile. Sterile instruments and surgical pack should be stored in a closed cabinet.

3. All sterilization devices used for sterilizing instruments for surgery must be tested for efficacy with **biological indicators.** There must be a log for efficacy testing of sterilization instruments completed on a semiannual basis. Use of alternative cold sterilants should be proposed in the IACUC protocol for review and approval.

4. All instruments, supplies, and devices that penetrate the skin or that will be implanted (needles, suture material, catheters, trocars, telemeters, etc.) must be sterilized by an approved method [see Table 1 below] prior to the initiation of surgery. Some catheter material cannot withstand the high temperature of autoclaving, e.g. polyethylene. Alternative options for sterilization of instruments include ethylene oxide (EtO), hydrogen peroxide vapor or cold sterilization (EtO and hydrogen peroxide vapor are not available at IUB). In the instance where surgery will be performed on multiple rodents, the surgery must start with an instrument pack that is sterilized then, instruments should be cleaned with sterile water, sterile saline or alcohol to lyse the RBC’s and remove debris and tips placed in the hot bead sterilizer. Allow appropriate cooling time of instruments before proceeding to use in next surgery. It is possible to use a “tips-only” technique. The instruments must start out fully sterilized, and for subsequent use please contact the Veterinary office to discuss further options in relation to “tips-only” or cold sterilization techniques.
**Recommended Instrument Sterilants**  
(Instruments should be cleaned of all gross debris prior to sterilization. Follow manufacturers’ recommended concentrations and contact times for all sterilizing agents.)

<table>
<thead>
<tr>
<th>AGENT</th>
<th>EXAMPLES</th>
<th>COMMENTS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Moist Heat (Steam)*</td>
<td>Autoclave</td>
<td>Effectiveness dependent upon temperature, pressure and time (e.g., 121°C for 15 minutes vs. 131°C for 3 minutes).</td>
</tr>
<tr>
<td>Dry Heat*</td>
<td>Hot Bead Sterilizer, Dry Chamber</td>
<td>Fast. Instruments must be clean prior to use and must be cooled before contacting tissue. Note that the hot bead sterilizer acts only on the tips of instruments and should therefore be used to re-sterilize the working end of instruments between surgeries, not as an initial means of instrument sterilization.</td>
</tr>
<tr>
<td>Gas Sterilization</td>
<td>Ethylene Oxide</td>
<td>Requires 30% or greater relative humidity for effectiveness against spores. Gas is irritating to tissue; all materials require safe airing time. May be used only after review and approval by the Environmental Health and Safety office.</td>
</tr>
<tr>
<td>H₂O₂ &amp; Peroxyacetic Acid</td>
<td>Sporklenz® (Full strength)</td>
<td>Broad spectrum. Sporcidal-fungicidal sterilant only with contact time of 11 hours or more. Instruments should be rinsed with sterile water or saline prior to tissue contact. Follow manufacturer’s precautions for safe handling, including use of personal protective equipment.</td>
</tr>
<tr>
<td>Chlorine</td>
<td>Chlorine Dioxide (Clidox®, Alcide®)</td>
<td>A minimum of 6 hours is required for sterilization. Presence of organic matter reduces activity. Must be freshly prepared (&lt;14 days). Instruments MUST be rinsed thoroughly with sterile water or saline prior to tissue contact. Follow manufacturer’s precautions for safe handling, including use of personal protective equipment.</td>
</tr>
<tr>
<td>Aldehydes</td>
<td>Formaldehyde (6% solution), Glutaraldehyde (Cidex®)</td>
<td>All aldehydes are sterilants only with many hours of contact. Corrosive and irritating. Consult with the Environmental Health and Safety office prior to using aldehydes for sterilization or disinfection. Instruments MUST be rinsed thoroughly with sterile water or saline prior to tissue contact.</td>
</tr>
<tr>
<td>Concentrated hydrogen peroxide</td>
<td>Plasma vapor sterilizer</td>
<td>Requires special equipment. Cycle takes about 50 minutes. Cannot be used for absorbable materials.</td>
</tr>
</tbody>
</table>

*Available at IUB

**B. Drapes:**

Drapes are used for wrapping the instrument packs and for creating the sterile field about the incision site. When opened, the wraps (sterile on the inside) become part of the sterile field. The use of drapes aids in covering the surgical animal to limit contamination from the animal’s fur or other body parts, especially if any suture material or catheters are to be used. Alternative materials such as sterile stockinets or Glad Press-N-Seal® may be used as drapes (contact LAR staff for details). Fenestrations can be created in the drape by cutting off the correct corner then unfolding the drape or cutting an appropriate size hole over the area of the incision.

**C. Surgical gloves:**

*Sterile surgical gloves must be worn* for all surgeries. Sterile surgical gloves are available commercially in different sizes and should fit snugly without causing numbness in the fingertips. Hypoallergenic gloves for surgeons with sensitive skin are also available. Gloves must be donned in a manner that prevents contamination of their exterior surfaces. Gloves should be changed if inadvertently contaminated or torn. The surgeon’s gloves are part of the sterile field and cannot touch anything outside of that field.

**D. Surgical wear:**

When performing surgery on rodents, a clean, close fitting **surgical mask**, and a clean surgical **scrub top, laboratory coat, or gown, hair cover and sterile surgical gloves** should be
considered the minimum of special surgical garb. This may be worn and need not be changed between rodents unless grossly contaminated. Use of safety glasses is highly recommended.

E. **Sutures/staples:**
Sterile, prepackaged suture material, with or without attached needles, is available commercially. Once opened, the suture length should be used on one animal only. Surgical wound clips or staples are an excellent alternative to sutures, especially in the skin. Wound clips must be sterilized prior to using. The loaded stapler is handled like the other instruments and the stapling technique is much faster than suturing. **Any skin suture or staples placed on an animal should be removed 7-10 days post-surgery,** unless scientifically justified and approved by the BIACUC.

### Surgical Location

1. Recovery rodent surgery may be performed in the laboratory, in a procedural room within an animal facility, or in a surgery suite. It must be **easily sanitized and not used for any other purpose during the time of surgery.**
2. The dedicated surgical area must be free of clutter and of adequate size to accommodate the animal, supplies, equipment and surgeon. The site chosen for animal preparation should be sufficiently distant from the surgical area to preclude contamination with any loose hair etc. Irrespective of the location, order and cleanliness are vital to successful aseptic technique.
3. The surgeon should plan in advance each step of the procedure, allocating space for induction of general anesthesia, the sterile field and placement of equipment and supplies within it, and recovery cages for animals. Particularly when multiple surgeries are performed at one session, there are advantages to performing surgery with 2 or more people.
4. The location selected for the sterile field must be disinfected prior to any surgery on a given day.

**Recommended Hard Surface Disinfectants (e.g., table tops, equipment)**
*(Follow manufacturers’ recommended concentrations and contact times for all disinfecting agents.)*

<table>
<thead>
<tr>
<th>AGENT</th>
<th>EXAMPLES</th>
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</tr>
</thead>
<tbody>
<tr>
<td><strong>Accelerated H₂O₂</strong></td>
<td>Rescue®</td>
<td>Broad spectrum disinfectant. Contact time of 5 min.</td>
</tr>
<tr>
<td><strong>H₂O₂ &amp; Peroxyacetic Acid</strong></td>
<td>Sporklenz®</td>
<td>Broad spectrum disinfectant. Requires contact time of ≥10 minutes. Follow manufacturer's precautions for safe handling, including use of personal protective equipment.</td>
</tr>
<tr>
<td>Chlorhexidine</td>
<td>Nolvasan® Solution, Hibiclens®</td>
<td>Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses.</td>
</tr>
<tr>
<td>Phenolics</td>
<td>LPH®, Vesphe®ne®, Lysol®</td>
<td>Less affected by organic material than other disinfectants.</td>
</tr>
<tr>
<td>Glutaraldehydes</td>
<td>Cidex®, Omnicide®, Banacide®</td>
<td>High-level disinfectants with 5-45 min. contact time; can be used as a sterilant with multiple hours contact time; exposure time limits set by Occupational Safety and Health Administration (OSHA).</td>
</tr>
<tr>
<td>Alcohols</td>
<td>40% ethyl alcohol, 85% isopropyl alcohol</td>
<td>Contact time required is 15 minutes. Contaminated surfaces take longer to disinfect. Remove gross contamination before using.</td>
</tr>
<tr>
<td>Chlorine</td>
<td>Sodium hypochlorite (Clorox- 10% solution), Chlorine dioxide (Cldox®, Alcide®)</td>
<td>Corrosive. Presence of organic matter reduces activity. Chlorine dioxide must be fresh (&lt;14 days old); kills vegetative organisms within 3 minutes of contact.</td>
</tr>
<tr>
<td>Quaternary Ammonium</td>
<td>Coverage®, TBQ®, Roccal®</td>
<td>Rapidly inactivated by organic matter. Compounds may support growth of gram-negative bacteria.</td>
</tr>
</tbody>
</table>
Animal Preparation

1. The animal should be **appropriately anesthetized** to a level that renders them unresponsive to external stimuli. An animal that is properly anesthetized for surgery does not flinch or vocalize at any time during the procedure. Depth of anesthesia can be determined by a toe pinch. If the animal does not respond to a toe pinch, it is under the appropriate surgical plane to conduct surgery. Many agents are available and appropriate for this purpose and should be recommended by a veterinarian. Please see LAR Drug Formulary for further details and contact LAR veterinary staff with any questions.

2. An **external heat source** should be provided for rodents during general anesthesia or sedation. It is vital that the animal's body temperature be maintained within acceptable limits as hypothermia is a significant cause of peri-surgical mortality in rodents.

<table>
<thead>
<tr>
<th>Species</th>
<th>Normal Body Temperature</th>
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<tbody>
<tr>
<td>Mouse</td>
<td>96.4°-99.7°F (35.8°-37.6°C)</td>
</tr>
<tr>
<td>Rat</td>
<td>96.6°-99.5°F (35.9°-37.5°C)</td>
</tr>
</tbody>
</table>

When anesthetized, the animal’s metabolic rate is depressed and body temperature promptly falls, owing to the high ratio of surface area to body mass. This temperature loss further slows the metabolism, extends the duration of the anesthetic agent(s), and can easily become life threatening. Maintenance of body temperature can be accomplished with a circulating water or warm air heating pad, warm water bottle, or microwavable heating device placed beneath the animal. Heat lamps and electrical heating pads are strongly discouraged.

3. A **lubricating ophthalmic ointment** should also be placed in an anesthetized animal’s eyes to prevent drying (Lacri-lube, Puralube ointment). Anesthetized animals do not produce a normal tear film and cannot blink to protect cornea. Ointment may need to be re-applied with longer procedures.

4. **Hair must be removed from the surgical site** for proper aseptic technique. Remove sufficient hair/fur to ensure that instruments will have no contact with the pelage during surgery.
   a. **Clipping:** Electrical clippers may be utilized on animals prior to performing approved procedures. The closeness of the hair clipped will be contingent upon the rationale for hair removal and the size of clippers used. Using clippers can be immediately followed or replaced by the use of a chemical depilatory agent for further hair removal.
   b. **Depilatory Creams:** Chemical depilatory hair removal agents may be used on animals designated for approved procedures. Depilatory creams (such as Nair®, Veet®, etc.) have been proven to be effective, atraumatic, as well as non-toxic. Depilation has also been associated with a significant reduction in skin surface bacteria. An advantage of using a depilatory agent is its ease of use in areas that are difficult to shave. Application of such products should be made in accordance with the manufacturer’s directives by placing a layer upon the area to be depilated for the designated time-frame. The depilatory and hair are removed by wiping the area with a water-moistened gauze pad or cloth. It is imperative that all traces of the depilatory cream be removed in order to avoid possible irritation from excessive exposure to the chemical agent. Although some animals may require a second brief treatment, the optimal time-frame of exposure for each strain can be determined by performing a test application on a smaller region, prior to the procedure.
   c. **Shaving:** Shaving with a razor may take place on animals before an approved procedure is performed. Shaving is becoming less common because it produces microscopic defects in skin
that have been associated with increasing the risk of site infections. However, when compared to alternative techniques such as chemical depilation, no statistical difference has been found between the methods. Topical analgesia may be provided for animals experiencing minor skin irritation related to shaving, clipping or depilation. Call the veterinarian to obtain advice on using analgesics.

5. Immediately prior to surgery, the **skin must be cleansed and disinfected**. This step creates part of the sterile field; therefore, care must be taken to avoid subsequent contamination. Caution must be exercised during these preparations not to drench the animal with excess fluid of any sort, especially alcohol. Excessive cooling of the animal's body may occur, which will prolong recovery times from the anesthetic and is potentially lethal. The surgical scrub should occur from the incision site spiraling outward, repeating the procedure at least 3 times using separate gauze or applicators so as not to contaminate the incision site during preparation. Surgical scrub may be used in an alternating fashion with sterile saline, applying each solution 3 times.

**Recommended Skin Disinfectants** (Note: Although it may appropriately be alternated with other agents, such as iodophor scrub, alcohol alone is not an adequate skin disinfectant. Also, the evaporation of alcohol can induce hypothermia in small animals and may reduce the efficacy of the other disinfectant.)

<table>
<thead>
<tr>
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<th>EXAMPLES</th>
<th>COMMENTS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorhexidine</td>
<td>Nolvasan® Scrub, Hibiclens®</td>
<td>Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses. Excellent for use on skin.</td>
</tr>
<tr>
<td>Warm, sterile saline, sterile water, or alcohol</td>
<td>Use as rinse solution</td>
<td>Since rodents can quickly and easily become hypothermic, the use of warm, sterile saline or sterile water is preferable over the use of alcohols that, upon evaporation, can quickly induce hypothermia.</td>
</tr>
</tbody>
</table>

6. The anesthetized animal should be secured with an appropriate method to prevent contamination of the surgical site. The animal should be positioned with the head and neck fully extended to ensure a patent airway.

7. **Sterile surgical drapes** are to be used to cover the animal’s body to provide a sterile field and to prevent contamination of the operative site and sterile surgical instruments. The animal must be carefully and **routinely monitored by visualization every couple of minutes** for normal vital parameters (e.g., heart rate, respiratory rate, body temperature, peripheral circulation) usually by visual observation and touch.

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**Surgeon Preparation**

1. After putting on the surgical garb, and prior to gloving and start of surgery, the surgeon should wash his/her hands thoroughly, preferably with microbicidal soap. The complete surgical scrub mandated for the aseptic operating rooms is not required for rodents. The surgeon's hands are dried with a clean towel.

2. Sterile surgical gloves must be donned in a fashion which avoids contamination of their exterior surfaces. Once the gloves are on, the surgeon may touch only within the sterile field; this means that all packs and supplies must be opened ahead of time. Gloves should be changed between
animals or disinfected if they become ripped or inadvertently contaminated by contact outside
the sterile field.
3. Safety glasses are optional.

Suturing and Suture Materials

The art of suturing entails the anatomically correct closure of the tissue layers incised during the
approach to the site of interest. The sutures placed in muscle, fascia, skin, etc. are meant to realign the
edges of these tissues such that rapid healing with minimal adhesions may occur.

Many materials are commercially available for use as sutures. Generally speaking, multifilament, braided
material (e.g. silk, Dexon, Vicryl) is used inside the animal, but is not appropriate for closing the skin. It
has good to excellent handling qualities and holds knots well but the “wicking” action of the
multifilament strand pulls fluids and microorganisms into the surgical wound and is not appropriate for
skin closure. Mono filament materials (e.g. nylon, Prolene) tend to be more difficult to handle and
requires more throws per knot to be secure, but lacks wicking action and is appropriate in the skin. For
suturing purposes, swaged-on (permanently attached) needles are preferable and are much easier to
use. Usually a cutting-edge needle is used to suture the skin. Monofilament, nonabsorbable sutures
placed in skin should be removed after healing has occurred, generally in 7-10 days.

Recommended Suture and Closure Materials (Note: Always use the smallest gauge suture material that
will perform adequately. Cutting or reverse cutting needles provide edges that will cut through dense, difficult to penetrate
tissue, such as skin. Taper point or round needles have no edges to cut through tissue and are used primarily for suturing easily
torn tissues, such as peritoneum or intestine.)

<table>
<thead>
<tr>
<th>MATERIAL</th>
<th>CHARACTERISTICS AND TYPICAL USES</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vicryl®, Dexon®</td>
<td>Multifilament. Absorbable in 60-90 days. Used for ligation or suturing where absorbable material is desirable.</td>
</tr>
<tr>
<td>PDS®, Maxon®</td>
<td>Monofilament. Absorbable in 6 months. Used for ligation or suturing where absorbable material is desirable, yet extended wound support is required.</td>
</tr>
<tr>
<td>Nylon</td>
<td>Nonabsorbable; inert. Used for general closure.</td>
</tr>
<tr>
<td>Silk</td>
<td>Multifilament. Nonabsorbable. Considered tissue reactive, and may wick organisms into the wound. Excellent handling. Is a preferred material for cardiovascular procedures.</td>
</tr>
<tr>
<td>Chromic Gut</td>
<td>Multifilament. Absorbable. Moderate tissue reactivity. rapid loss of strength. For internal use only.</td>
</tr>
<tr>
<td>Stainless Steel Wound Clips or Staples</td>
<td>Nonabsorbable. Requires special instrument for removal from skin. Must not be clamped too tight.</td>
</tr>
<tr>
<td>Nexaband®, Vetbond or other Tissue Adhesives</td>
<td>Absorbable and fast. Slightly less tensile strength than standard suture materials.</td>
</tr>
</tbody>
</table>

Required Documentation

The Principal Investigator (PI)/surgeon is responsible for maintaining accurate records regarding surgical
procedure and perioperative care. Group or individual records that detail procedures, drugs
administered, dates, personnel, pre- and post-surgical condition of the animal, identification of the
animal, body weight, identification of the surgeon and documentation of 7-10 days of postoperative
monitoring must be kept for animals that have undergone surgery. Intra- and post-operative
documentation is required from laboratories that perform recovery surgical procedures on rodents.
Surgical Record Forms are attached to this document as a recommended method for recording this
information. Records of active cases must be readily available to the personnel involved in
post-surgical monitoring, the veterinary staff, the IACUC, and federal regulatory officials.
Surgical records should be maintained in or near animal housing areas for active cases. Only records of live animals present in housing areas must be readily available. Surgical records are to be maintained for 3 years following the death of the animal or following the expiration of the related protocol.

Post-Operative Monitoring

Animals recovering from general anesthesia following surgery require careful and consistent monitoring by trained personnel. After surgery, animals should be placed in a warm, dry area and monitored until they have recovered from anesthesia and are ambulatory. External heat sources such as a warm water or air blanket can provide the needed external heat source. Ideally, rodents should be housed individually until they are ambulatory to prevent cannibalism or injury to other rodents. The recovery area should be near the surgery area to allow observation of recovering animals while work is occurring. Animals taking longer than expected to recover should be gently handled and examined, and their position changed in the recovery cage.

Animals that have recovered from surgery (can ambulate in the cage) can be placed back in their housing room with appropriate identification that indicates the animal had surgery and the date of the surgical procedure. Appropriate postoperative monitoring of surgical animals should continue daily for at least 7-10 days after surgery with sutures or staples removed at the end of the observation period. The animal's incision site should be observed for signs of redness, discharge or dehiscence. Appetite, attitude, hydration and temperature should be noted as an indication of recovery from the surgery. Supplemental fluids, analgesics and antibiotics should be administered post-operatively as needed and approved in the animal use protocol. Special diets, housing, and environmental conditions (e.g., temperature, humidity) should be considered to maximize the rate of healing. Surgical information should be maintained, including but not limited to the type of procedure, date performed, any problems encountered, and the type/dose/route of analgesic administered as appropriate. Monitoring of the animal’s progress, administration of medications, and management of the surgical incision up to the time of suture removal are to occur daily. If there are any questions about the recovery of a postoperative animal, a veterinarian should be consulted.

Researchers may contact trained LAR staff to perform surgery or provide post-anesthetic/post-surgical monitoring for a fee. At least 5 business days advanced notice should be provided. Please be aware that the IACUC and LAR are required to ensure that animals are properly cared for and that medical records are appropriately maintained. If it is discovered that appropriate care is not being provided to an animal, LAR will provide this care. All matters will then be investigated by the IACUC.

Non-survival Rodent Surgery

Consideration should be given to applying the above Guidelines when performing non-survival surgery that lasts longer than a few hours, in order to decrease the chance of introducing bacteria that may colonize the surgical site or equipment and produce untoward effects within a short period of time. At a minimum, per the Guide, the surgical site should be clipped, the surgical bench and instruments should be clean, and the surgeon should wear gloves.
Quick Reference Guide
For Rodents Receiving Anesthesia and/or Surgery

PREPARATION OF THE SURGERY AREA
1. Area is a room or portion of a room that is easily sanitized and not used for any other purpose during the time of surgery - keep it free of clutter.
2. Clean or disinfect the surface upon which surgery will be performed with one of the environmental disinfectants listed in the document above under "Recommended Hard Surface Disinfectants".
3. Use an LAR surgical monitoring form or one approved by the IACUC to record information on animals undergoing surgery.

PREPARATION OF SURGICAL INSTRUMENTS
1. Sterilize surgical instruments with one of the sterilizing agents listed above in "Recommended Instrument Sterilants".
2. For multiple surgeries performed during a single session:
   - Start with at least 2 sets of sterile instruments.
   - Between animals, clean and disinfect instruments with one of the high-level instrument disinfectants listed in "Recommended Instrument Sterilants", or clean and place in hot bead sterilizer for 20 sec.

PREPARATION OF THE ANIMAL
1. To keep the animal warm, place it on a clean absorbent surface; placement of a temperature-controlled heating blanket, warm water bottle, or equivalent under the pad is strongly recommended.
2. Put an ophthalmic ointment in the anesthetized animal's eyes to prevent drying.
3. Prior to taking the animal to the surgery area, remove hair with animal clippers, razor, or depilatory cream; vacuum or otherwise remove the loose hair.
4. Clean and disinfect the animal's skin by scrubbing with one of the skin disinfectants listed in "Recommended Skin Disinfectants" alternating with sterile saline, sterile water or alcohol.
5. Cover the animal with a surgical drape.

PREPARATION OF THE SURGEON
1. Wash hands before surgery with disinfectant soap.
2. Wear a mask, sterile gloves, clean scrub top, lab coat or gown, and hair cover; if contaminated or torn, a new pair of gloves should be worn for each animal.
3. Safety glasses are recommended.

POSTOPERATIVE CARE
Immediately after surgery:
1. Check the animal regularly at least every 15 minutes until it is fully ambulatory and record observations on the individual or group surgery record.
2. To minimize hypothermia, place the animal in a warm room on a bedded or padded surface; provide additional warmth with something such as a warm water blanket or warm water bottle.
3. To help prevent dehydration, warmed fluids may be administered per IACUC approved protocol.
4. To avoid cannibalism by cage mates, house rodents individually or in small groups if under constant supervision until they are fully ambulatory. Place in home cage in housing room only after ambulatory.
5. After the immediate postoperative period, check the animal at least once a day to assure freedom from postoperative complications.
6. Record preoperative and postoperative monitoring on appropriate forms that are available for review by the LAR staff or regulatory personnel until the animal is fully recovered and incision has healed, about 7-10 days after surgery.
7. Contact an LAR veterinarian or veterinary technician (x52356) if the animal or surgical wound appears abnormal.
Sanctions

Failure to comply with IACUC policies may result in noncompliance reports to the Institutional Official, the Office of Laboratory Animal Welfare (OLAW), the U.S. Department of Agriculture (USDA), and/or the suspension of animal use privileges. In addition, the availability of sponsored research funds may be affected when an investigator is found to be in violation of these policies.

Additional Contacts

<table>
<thead>
<tr>
<th>Subject</th>
<th>Contact</th>
<th>Phone</th>
<th>Email</th>
</tr>
</thead>
<tbody>
<tr>
<td>Veterinary Concerns</td>
<td>LAR Veterinarians</td>
<td>855-2356</td>
<td><a href="mailto:lar@indiana.edu">lar@indiana.edu</a></td>
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<td>Policy</td>
<td>IACUC Manager</td>
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<td><a href="mailto:biacuc@indiana.edu">biacuc@indiana.edu</a></td>
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</table>

References