

Policy on the Rodent Survival Surgery

Date Reviewed: 12/11/19

1. Facility

- A. A dedicated facility for rodent surgery is not required. A rodent surgical area can be a room or portion of a room that is easily sanitized. The immediate surgical area must not be used for other purposes during the time of surgery.
- B. Surgery must be conducted on a clean, uncluttered lab bench or table. The surface of the lab bench or table must be impervious to liquids. The work surface must be wiped with disinfectant before and after use or covered with a clean drape.
- C. The surgery area MUST be separate from the area where hair is removed from the animal

2. Training

Professional and technical personnel and students who perform anesthesia, analgesia, and surgery must be trained to accomplish these tasks in a humane and scientifically acceptable manner before any surgery can take place. The UCSC veterinary staff is available to provide assistance with, or training in, aseptic and surgical techniques and the proper administration of anesthesia and analgesia.

3. Instruments

A. Instrument Preparation

All instruments must be cleaned and sterilized prior to use. First, all instruments must be cleaned of any debris by hand washing or by mechanical washer/sterilizer. Then, prior to surgery, the instruments must be sterilized using one of the following methods. The method of choice may be determined by the procedure, the delicacy of the surgical instruments or the devices being used. Steam autoclaving is the preferred method.

- 1. Heat Sterilization
 - a. Steam Autoclave: The instruments must be placed in a specially designed pack or wrapped in sterile drapes or cloths, and secured with a thermo-sensitive tape. The use of such tape provides some indication that the autoclave procedure was effective. Instruments must be autoclaved at 121oC for 21 minutes in a vacuum autoclave. Different times are required for gravity autoclaves. Once autoclaved, packs or wrapped instruments must be stored in closed cabinets or plastic bags. Autoclaved items must have a standard indicator to prove complete sterilization. Wrapped autoclaved items must be clearly labeled with the date of sterilization or expiration date. Items that are autoclaved in cloth wraps expire 6 months after

autoclaving. Items that are autoclaved in plastic packs expire 1 year from the date of autoclaving.

b. Flash Steam: Used to sterilize articles intended to be used immediately. The temperature must reach 132oC for three to five minutes.

2. Cold (Chemical) Sterilization

Effective and proper use of chemical sterilization depends on many factors, including the use of chemicals classified as sterilants (not disinfectants), physical properties of the item(s) being sterilized (i.e., smooth, impervious to moisture, clean) and assurance of proper exposure. Chemical sterilants have finite shelf lives and must be used, depending on the agent, within one to four weeks depending on the agent (follow label directions).

Furthermore, the solutions must be protected from contamination. Effective cold sterilization requires thorough cleaning of instruments prior to processing because blood and organic debris may inactivate chemical germicides and/or shield microorganisms from the sterilization process. Clean Rubbermaid-type containers with secure lids or stainless-steel instrument trays and lids are recommended for cold sterilization procedures and instrument storage. Sterile water or saline must be used to rinse the instruments, implants and tubing (inside and outside) prior to use to avoid tissue damage to the animals. The following are acceptable chemical sterilants:

- a. Alcide® Active ingredient: Chorine Sodium Chlorite 1.37%. Exposure time must exceed 6 hours. Shelf life is 14 days.
- b. Cetylcide-G: Active ingredient: 3.2% denatured glutaraldehyde. Exposure time of 20 minutes will kill bacteria, fungi and many viruses, but requires extensive incubation to kill bacterial spores. Full sterilization requires 10 hours. Shelf life is 28 days.
- c. Cidex® Active ingredient: 2% glutaraldehyde. Exposure time must exceed 10 hours for sterilization. Cidex comes in two formulations, Cidex and Cidex-7 (long-life). The shelf life of activated Cidex is 14 days and of activated Cidex-7 is 28 days.
- d. Endospore® Active ingredient: stabilized hydrogen peroxide 6%. Not acceptable for metallic items.
- e. Sporicidin® Active ingredient (activator + buffer): phenol 7.05%, glutaraldehyde 2%, and sodium phenate 1.2%. Exposure time must exceed 6.75 hours for sterilization. Shelf life is 28 days.
- f. Ethylene Oxide Gas: This is only used for instruments that will be damaged by heat or steam sterilization. This process is toxic, expensive and is regulated by federal law. Plastic, silicon and polyethylene catheters may be sterilized with ethylene oxide gas on the cool cycle.
- g. Volatile Hydrogen Peroxide (VHP): safe and ideal for most applications. Requires an expensive generator.

B. Rubber Tubing

The following methods can be used to sterilize rubber tubing

- 1. Heat sterilization
- 2. Ethylene oxide gas
- 3. 6% hydrogen peroxide solution
- 4. VHP (hydrogen peroxide)

C. Multiple Surgeries

If multiple surgeries are to be performed on different animals, then previously sterilized instruments can be "quick"- disinfected, using a glass bead sterilizer (at least 15 sec), 70% alcohol (10 minutes), or glutaraldehyde (10 min). However, instruments should be thoroughly clean of blood or tissue prior to sterilization. No more than five successive surgeries can use instruments "quick"-disinfected as described above.

- 1. Sterile (Hot) Bead Sterilizer: This instrument will sterilize the tips of metal instruments in 15 seconds. However, the beads should be clean. Only clean, cooled instruments may be used on the animals. Instruments must also be of appropriate size for the unit. This type of sterilization is ideal for multiple cage surgeries.
 - NOTE: Most sterile bead sterilizers take thirty minutes to heat.
 - NOTE: This method of sterilization may not be used for the initial sterilization of instruments; it is only appropriate when performing 5 or fewer surgeries using a single pack.
- 2. Chemical disinfectants: Instruments soaked in chemical disinfectants must be rinsed in sterile water or saline before use on animals.

4. Anesthesia and Analgesia Selection

Contact the Institutional Veterinarian for recommendations for appropriate anesthetics and/or analgesics for the species you are using.

The use of a single analgesic agent or combination will depend on the procedure performed. This table provides some guidelines for determining the expected degree of pain associated with various surgical procedures. For specific advice, please consult the Institutional Veterinarian.

SURGERY TYPE	ANALGESIC	DURATION OF TREATMENT
Subcutaneous incision	NSAID or opioid	Pre-emptive + 1 dose
Open abdomen	NSAID and opioid	Pre-emptive + 48 hours
Open thorax	NSAID and opioid and/or local	Pre-emptive + 48 hours
Musculoskeletal manipulation (e.g., fracture, muscle resection)	NSAID and/or opioid and/ or local	Pre-emptive + 48 hours



Open cranium	NSAID and/or opioid and/ or local	Pre-emptive + 48 hours
Implant or device placement (e.g., indwelling catheter)	NSAID and/or local	Pre-emptive + 24 hours

It is important to realize that none of the analgesics work immediately, so it is preferred that analgesics are provided pre-emptively to the painful event. Generally, this can be accomplished by administering the analgesics at the time of anesthetic induction or one – two hours before the surgical procedure. The specific analgesic choice and duration of administration to use is based on the severity of pain expected. These choices listed are not necessarily interchangeable. Please consult with the veterinary staff for additional guidance. Analgesics may be given preemptively (preferred), intra-operatively to reduce inhalant requirements and provide additional analgesia, and post-operatively.

Mouse

- Buprenorphine 0.05-2.0mg/kg SQ every 6-12 hours SQ, IP, or IM
- Buprenorphine 0.05-2.0mg/kg every 6-12 hours SQ or IM + Carprofen 5mg/kg q 6-8 hours
- Buprenorphine 0.05-2.0mg/kg every 6-12 hours SQ or IM + Meloxicam 1-2mg/kg q 24 hours
- Extended release buprenorphine 1.0 3.25 mg/kg SQ once.
- Carprofen 5-10mg/kg PO or SQ q 6-8 hours; can be combined with opioids
- Meloxicam 1.0-2.0mg/kg SQ, IP daily; can be combined with opioids
- Local: lidocaine, lidocaine/bupivicaine, lidocaine patch, bupivicaine

Rat

- Buprenorphine 0.01-0.05mg/kg SQ or IM every 8-12 hours
- Buprenorphine 0.01-0.05mg/kg SQ or IM every 8-12 hours + Carprofen 5mg/kg q 6-8 hours
- Buprenorphine 0.01-0.05mg/kg SQ or IM every 8-12 hours + Meloxicam 1-2 mg/kg once daily
- Extended release buprenorphine 1.0 3.25 mg/kg SQ once. Carprofen 5-10mg/kg orally or SQ q 6-8 hours; can be combined with opioids
- Meloxicam, 1.0-3.0mg/kg PO, SQ, IP daily; can be combined with opioids
- Local: lidocaine, lidocaine/bupivicaine, lidocaine patch, bupivacaine



5. Preparation of the Animal

- A. The animal must be anesthetized with a suitable anesthetic using the doses and procedure approved by the IACUC.
- B. An ophthalmic lubricant must be applied to the eyes to prevent corneal drying.
- C. Hair must be removed from the incision site with clippers, appropriate razor, and/or hair removal product (i.e., Nair) applied as directed and thoroughly rinsed off to prevent continual residue action.
- D. Skin Preparation: The bare skin at the incision site must be thoroughly scrubbed with a surgical antiseptic agent to disinfect the skin and create a sterile field around the incision site. Starting in the middle, and continuing in an outward spiral, apply the scrub at least three times alternating each scrub with 70% isopropyl or ethyl alcohol, sterile water or saline.

Note: Copious application of topical alcohol in rodents will soak the animal and lead to hypothermia. The use of cotton tip applicators are ideal during the skin preparation process.

These surgical antiseptic agents may be used:

- Povidone iodine scrub: A good choice for a surgical preparation with a broad spectrum of activity, including Mycobacterium. Antiseptic activity is rapid and persistent if not removed.
- 2. Chlorhexidine scrub: The 4% aqueous preparation effectively cleans the skin with a rapid onset of activity and a broad spectrum of activity with minimal loss of antiseptic activity.

NOTE: A scrub is different than a solution. A scrub contains a soap, and therefore has cleaning properties that a solution does not have. Scrubs are not to be mixed or diluted with water.

Antiseptic agents must be rinsed from the skin with sterile water, sterile saline or alcohol prior to surgery.

6. Preparation by Surgeon

- A. Hands must be washed with an antiseptic soap or a surgical detergent/scrub (iodophors or chlorhexidine) and rinsed with water. <u>Sterile</u> surgical gloves must be worn. Non-sterile exam gloves are not acceptable.
- B. A surgical mask must be worn to prevent contamination of the surgical field.
- C. Gowns and surgical bonnets are required to maintain a sterile surgical field. The sleeves of garments must not be allowed to come in contact with sterile surfaces (e.g., gloves, the animal, etc.).
- D. A new pair of <u>sterile</u> surgical gloves must be used for each animal. Alternatively, surgeons may wipe their gloves for 30 seconds with sterile gauze pads soaked in



- 70% alcohol, or with chlorine dioxide for 3 minutes. Gloves must be wiped with 70% alcohol after the 3-minute chlorine dioxide application.
- E. If working alone, the surgeon must have the animal anesthetized and positioned prior to gloving.
- F. If the instruments are in a sterile pack, the first layer of the double-wrapped instrument pack must be opened before gloving.
- G. For survival surgery, the surgical site must be covered with a sterile drape after the surgeon has donned **sterile** gloves.

7. Intraoperative Monitoring

- A. The animal must be monitored carefully during the surgical procedure. Specifically, the animal's respiratory rate and characteristic response to noxious stimuli (e.g., toe pinch, and when possible the heart rate and body temperature) will be monitored.
- B. The surgical team must be trained by experienced personnel to be able to respond to the most common emergencies associated with the type of procedure being performed.

8. Post-Surgical Care

- A. Post-surgical care must include observing the animal to ensure uneventful recovery from anesthesia and surgery, administering analgesics, providing adequate care to surgical incisions and maintaining appropriate medical records.
- B. **Administration of analgesia is required**, except when specific <u>IACUC approval</u> has been granted.
- C. To prevent hypothermia, place the animal(s) in a warm room or cage. Do not place the recovering animal directly on the cage bedding. To prevent suffocation of the animal, It is recommended to recover the animal in a cage without bedding. The cage may be placed on a bedded or padded surface and supplied with extra bedding or supplemental heat. Water-circulating heating blankets are recommended instead of electrical heat sources. Heating blankets must be covered to avoid direct contact with the animal. Do not place animals directly under a heating lamp, as it may cause thermal burns. Heat lamps are not allowed for use with rodents.
- D. Dehydration can be ameliorated by the administration of appropriate fluid therapy. Initially this may be done by giving 1 to 2 ml of warm (approximately 37oC) sterile fluids (0.9% NaCl or Lactate-Ringers Solution) per 100 gm of body weight by subcutaneous or intraperitoneal injection. If blood loss occurred during the surgical procedure or if the animal is slow to recover from the anesthetic, additional fluids may be necessary.
- E. During the recovery process, animals must be monitored continually until they gain the righting reflex.

- F. If recovery from the anesthetic will be prolonged (i.e., over one hour), the animal must be rotated from side to side every 15-30 minutes to minimize atelectasis (collapse) of the lungs. This practice must be continued until the animal regains the righting reflex.
- G. Post-surgical care of animals must be evaluated daily for at least five days by a member of the principal investigator's staff to whom post-operative care has been delegated. Animals must be monitored for evidence of excessive inflammation at the incision site, suture dehiscence (incision line failure or separation), infection, behavioral abnormalities indicative of illness (anorexia, listlessness, lethargy, dehydration, ruffled coating, lack of movement, weight loss greater than 10%). If evidence of wound infection or illness is noted, then the veterinarian must be contacted for evaluation and treatment, or the animal must be euthanized as soon as possible.
- H. External sutures, staples, and wound clips must be removed 10-14 days after surgery, unless otherwise approved in the protocol or approved by the veterinarian.
- I. If infections or complications occur, the vivarium manager and the veterinarian must be notified immediately.

9. Surgical Records

- A. "Surgical Record" must be completed immediately after the surgical procedure is performed. Records may be somewhat abbreviated and can be included as part of the research data collected, but must also be available for review.
- B. Records must identify:
 - 1. The type of surgical procedure performed
 - 2. The date of the procedure
 - 3. The person who performed the procedure (or initials)Information on all drug administration (including anesthesia and analgesia) and peri-operative monitoring
 - Records must be maintained by the laboratory (lab notebook).
 - 5. This information must be available for review by regulatory bodies, including the IACUC.

10. Suture Selection

Close surgical wounds using appropriate techniques and materials. The following table is a guide to the types of sutures that are available.

Suture	Characteristics and Frequent Uses
Vicryl®, Dexon®	Absorbable; 60-90 days. Ligate or suture tissues where an absorbable suture is desirable.
PDS®, Maxon®	Absorbable; 6 months. Ligate or suture tissues especially where an



	absorbable suture and extended wound support is desirable.
Prolene [®]	Nonabsorbable, Inert. General skin closure.
Nylon	Nonabsorbable. Inert. General skin closure.
Silk	Nonabsorbable. (Caution: Tissue reactive and may wick microorganisms into the wound). Excellent handling. Preferred for cardiovascular procedures. Must not be used to suture skin.
Chromic Gut	Absorbable. Versatile material. Because gut is highly reactive to tissues, its use is discouraged.
Stainless Steel Wound Clips, Staples	Nonabsorbable. General skin closure.
Cyanoacrylate surgical glue	Generally used in addition to skin sutures or incisions less than 1 cm in length. Note that many rodents will rip the glue out, along with large areas of skin, making it difficult to close the incision. Thus, surgical glue is not recommended.

11. Exceptions

All planned deviations from this policy must be approved by the IACUC prior to the performance of the surgical procedure. Emergency situations that involve deviations from IACUC-approved procedures must be reported to the veterinarian and IACUC committee within one week of its occurrence.