UBC Animal Care Guidelines
SOP: ACC-02-2017 Rodent Survival surgery
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This SOP will be used for: Full aseptic surgery ☐
Sterile tip surgery ☐
Both full aseptic and sterile tip surgery ☐

Rodent Survival Surgery SOP

1. Purpose

This Standard Operating Procedure (SOP) describes methods to be followed for rodent survival surgery. This SOP follows the UBC Policy and CCAC guidelines for the survival surgery of rodents.

2. References:

Associated Guidelines and SOP's can be found on the ACC SOP/Guidelines website:
https://animalcare.ubc.ca/planning-your-research/sops-guidelines

TECH 10 - IP injection in mice and rats

TECH 11 – Subcutaneous injection in mice and rats

SOP ACC-2012-01 Rodent Anesthesia

Rodent Anesthesia and Analgesia Formulary and General Drug Information

Surgical Class and Analgesic Guidelines

NSAID Analgesia for Rodents: Ketoprofen (Anafen)

NSAID Analgesia for Rodents: Meloxicam

Opioid Analgesia for Rodents: Buprenorphine

Local Analgesia/Anesthesia for Rodents: Lidocaine, Bupivacaine, Ropivacaine

Policy # 16 Survival Surgery of Rodents

Policy # 17 Monitoring of Animals Used for Research, Teaching and Testing

Recommended suture material and sizes for closing surgical incisions in rats and mice

	Type	Size	Closure Pattern
Abdominal wall/muscle	Absorbable (Vicryl, PDS, Monocryl)	Rats (4-0 to 6-0) Mice (5-0 to 6-0)	Simple continuous
Subcutaneous tissue	Absorbable (Vicryl, PDS, Monocryl)	Rats (4-0 to 6-0) Mice (5-0 to 6-0)	Simple continuous
Skin	Non-absorbable, monofilament (Prolene, Nylon)	Rats (4-0 to 6-0) Mice (5-0 to 6-0)	Simple interrupted

*Absorbable suture and a simple continuous pattern can be used for skin suturing if a subcuticular (buried) suture pattern is used or if a monofilament absorbable suture such as Monocryl is used.

**A useful suture needle is a swaged on RB-1 taper needle for most rodent tissues. In some cases, rat skin is thicker and tougher than mouse skin and may require a swaged on reverse cutting needle (1/2 circle).

3. Responsibility

3.1. Principal Investigators (PIs) and their research staff, animal care staff and veterinary care staff.

3.2. All animal users performing surgery on rodents must have successfully completed the UBC Animal Care Centre (or equivalent) the UBC (or equivalent) Introduction to Working with Rodents in Research, Restraint, SQ and IP injection courses, Introduction to Rodent Anesthesia and the Introduction to Rodent Aseptic Surgery courses.

4. Introduction

4.1. **Aseptic technique** is a set of specific practices and procedures performed under carefully controlled conditions with the goal of minimizing contamination by potential pathogens.

4.2. In order to comply with Policy # 16 Survival Surgery of Rodents, steps must be followed to prepare the surgical area, animal, surgeon, and surgical instruments, supplies and/or implants.
4.3. Though rodents may survive a surgical procedure that is not done aseptically, major physiological changes will occur as the animal's immune system fights infection and these can significantly impact animal welfare and research results.
4.4. Though not required, aseptic techniques for non-recovery surgical procedures (especially those that are longer than 1 hour) are highly recommended in order to prevent physiological changes that confound research results.
4.5. The goals of a successful surgery are: maintenance of asepsis, minimizing surgery time, minimizing tissue trauma by gentle tissue handling, and maintenance of normal rodent physiological parameters for the given surgical model.
4.6. Never leave an anesthetized animal unattended. Monitor animal until it has recovered from anesthesia enough to be moving around the cage normally.
4.7. Monitoring of and record keeping for post surgical animals should occur as described in the Surgical Class and Analgesic Guidelines document, Policy # 17 Monitoring of Animals Used for Research, Teaching and Testing and the approved Animal Care Protocol and must continue until the surgical incision is healed, sutures are removed, pre-surgical weight recovers or remains stable and the animal has recovered to the point that is normal for the given surgical model (see Surgical Class and Analgesia Guidelines).
4.8. Suture material must not be reused from animal to animal.

5. Materials

5.1. Safe heat source (i.e.: homeothermic feedback blanket, warm-water circulating pad, infrared heater. Electrical heating pads can be used but the surface temperature must be measured before and during use to prevent burns).
5.2. Corneal protectant (i.e.: lacrilube, isoptotears, TearGel, etc)
5.3. Accurate animal weigh scale
5.4. Calculator
5.5. 70% Isopropyl alcohol
5.6. Disinfectant for surfaces
5.7. Paper towels
5.8. Warmed SQ fluids (i.e.: sterile 0.9% saline or sterile lactated ringer's solution) if anesthesia/surgery is expected to last more than 10 minutes.
5.9. Appropriately sized needles and syringes for size of animals (See TECH 11 SQ injection in mice and rats and TECH 10- IP injection in mice and rats).
5.10. Warmed recovery cage with paper towel covering bottom. No loose bedding should be present for animals not fully recovered from anesthetic.
5.11. Easily accessible food and water or gel water replacement (e.g.: Transgel, Recovery Gel, Hydrogel, etc).
5.12. Appropriate analgesics (see Policy # 16 Survival Surgery of Rodents, Surgical Class and Analgesia Guidelines and Rodent Anesthesia and Analgesia Formulary and General Drug Information).
5.13. Appropriate anesthetics (see SOP ACC-2012-01 – Rodent Anesthesia)
5.14. Cage flags, surgical log and/or monitoring sheets (see Policy # 17 Monitoring of Animals Used for Research, Teaching and Testing).
5.15. Appropriately sized rectal thermometer if anesthesia is expected to last more than 10 minutes (typically, if the probe diameter is the smaller than the diameter of the animal's normal fecal pellet diameter, it can be safely used)
5.16. Additional patient monitoring equipment if available – pulse oximeter, capnograph (CO_2 monitor)
5.17. Electric fur clipper (with size 40-50 blade)
5.18. Gauze
5.19. Cotton tipped applicators
5.20. Chlorhexidine (Hibitane) 2% surgical scrub or equivalent (i.e. Betadine scrub)
5.21. Water resistant surgical pack wraps (Cloth wrappers, disposable Steri-wraps or Steri-pouches)

5.22. Chemical sterilization indicators
5.23. Autoclave tape
5.24. Sterilized (via steam autoclave) surgical instruments appropriate for planned surgery
5.25. Sterile gauze
5.26. Sterile cotton tipped applicators
5.27. Sterile drape with appropriately sized fenestration (hole) for surgical incision
5.28. Hot bead sterilizer (if sterile tip surgery is planned).
5.29. Bouffants (hair covers)
5.30. Surgical masks or N95 masks
5.31. Non-sterile exam gloves
5.32. Clean lab coat, scrub top or gown
5.33. Sterile towels or sterile paper towels
5.34. Sterile surgical gloves of the appropriate size for surgeon (if full sterile technique is being used)
5.35. Sterile suture material of appropriate size and type for surgery (see recommendations above).
5.36. Animal surgical log listing type of surgery, anesthetics and analgesics given
5.37. Cage flag cards indicating type of surgery, anesthetics, analgesics given
5.38. Monitoring sheets with duration and frequency of monitoring for parameters listed on approved animal care protocol

6. Preparation of surgical instruments and sterile supplies (typically done a day or two before planned surgery)

6.1. Clean all surgical instruments needed for surgery and wrap in chosen surgical pack for autoclaving. Include a chemical sterilization indicator in each pack. Place autoclave tape on outer surface of each pack.
6.2. Label each pack with date of autoclaving, name of person preparing the pack (and contents if desired).
6.3. In packs, include any required additional sterile supplies such as drapes, gauze, cotton-tipped applicators, sterile tip fields, bowls, etc.
6.4. Prepare packs for autoclave and set autoclave parameters to the necessary time, pressure and temperature as required to achieve full sterilization of the packs
6.4.1. A biological indicator should be placed in the center of the largest pack in the centre of the load once a month to ensure the autoclave parameters chosen are achieving sterility.
6.5. Remove dried packs from autoclave, check that autoclave tape indicates packs have been processed, and place in an area where the packs will not get wet or dirty.

7. Preparation of surgical and prep areas (performed right before planned surgery)

7.1. Choose location for surgery area – it should be away from busy areas (such as doorways), easily cleaned and disinfected, and not directly under an unfiltered air supply duct.
7.2. Remove all unnecessary equipment and clutter from area and clean surfaces to remove dust, fur, bedding, etc.
7.3. Disinfect surfaces in surgical and prep areas with a disinfectant (i.e. Virkon, Percept, etc) and ensure proper contact time (refer to manufacturer's recommendations).
7.4. The surgical and prep areas should be cleaned and disinfected before and after each surgical procedure to remove blood, urine, feces, hair, etc.
7.5. Bring required support and monitoring equipment to surgical and prep areas (i.e. heating source, syringes, needles, clippers, etc).
7.6. Bring sterile surgical packs and supplies to surgery area but do not open yet.
7.7. Turn on hot bead sterilizer if using sterile tip technique

8. Preparation of Animal

8.1. Put on non-sterile exam gloves, surgical mask, hair bouffant and a clean lab coat or scrub top
8.2. Calculate and draw up required anesthetics, analgesics and fluids. Record all drugs and volumes on Surgical log.
8.3. Anesthetize the animal according to SOP ACC-2012-01– Rodent Anesthesia in the designated prep area on a safe heat source.
8.4. Once immobile, place animal on heat support, apply corneal protectant in both eyes, and administer required analgesics (see above references) and SQ fluids.

8.5. Remove hair from planned surgical site with fur clippers being careful not to cut the skin with the clippers. Shave at least 1 cm perimeter around planned surgical site for mice and 2cm perimeter for rats.

8.6. Remove all loose hair (using gauze or a small handheld vacuum)

8.7. Clean the skin at the planned surgical site with 2% chlorhexidine soap (or equivalent) using gauze (rats) or cotton-tipped applicators (mice or heads of rats). Start cleaning directly over where the planned incision will be and extend outwards without touching the center again. Take care not to wet or soak the animal as this will lead to heat loss.

8.8. Remove soap with 70% isopropanol on a gauze or cotton tipped applicator in the same pattern as the soap was applied (centre to periphery). DO NOT spray animal with the alcohol or get it near the eyes.

8.9. Discard used gauze and cotton tipped applicators after each application of soap or alcohol.

8.10. Administer local anesthetic (see Local Analgesia/Anesthesia for Rodents) at the planned surgical incision site.

8.11. Repeat the soap application and alcohol removal procedure two more times

8.12. Ensure all loose hair is removed from the animal and bring animal to the prepared surgical area and continue supportive care (heat, oxygen, inhalational anesthetic (if using).

8.13. Lubricate and place rectal thermometer and any other monitoring equipment (pulse oximeter, capnograph, etc).

8.14. Ensure animal is positioned correctly for the surgery and check breathing, color of extremities, temperature, and surgical level of anesthesia (rear toe pinch withdrawal)

8.15. Adjust anesthesia level if required (adjust isoflurane % or inject an additional small dose of injectable anesthetic being careful not to exceed therapeutic index).

8.16. Change gloves to a new pair of exam gloves

9. Initial preparation of Sterile Field

9.1. Carefully open the sterile instruments close to the anesthetized patient (within easy reach) without touching any inner surface of the pack.

9.2. Aseptically add all required sterile supplies to the sterile field by opening the outer package and letting them drop a short distance onto the sterile field (i.e.: scalpel blades, suture material, gauze, cotton tipped applicators, syringes, bowl to hold sterile saline or LRS, etc)

9.3. Draw up required sterile saline or LRS in a syringe and add fluids to sterile bowl (this is used to keep exposed tissues moist during surgery)

9.3.1. Ensure that they do not touch any non-sterile surfaces and the person does not touch any sterile surfaces.

9.4. Open outer package of sterile surgical gloves without contaminating inner surface

9.5. Check animal's breathing, temperature, color and anesthetic depth – make any required adjustments to support animal and maintain a surgical plane of anesthesia.

10. Preparation of the surgeon

10.1. Remove exam gloves and wash hands and arms well (up to elbows if exposed) with disinfectant soap (i.e.: Chlorhexidine).

10.1.1. Keep hands raised above elbows to prevent "dirty" water from running down arms and contaminating hands

10.1.2. Pay particular attention to underneath fingernails, between fingers, palms and wrist areas

10.1.3. A good hand wash should take about 90 seconds to complete

10.1.4. Dry hands with a sterile towel or paper towels

10.2. If wearing a sterile surgical gown, put this on

10.3. Put on sterile surgical gloves without touching outer surface if using full sterile technique, otherwise put on a clean pair of exam gloves.

10.3.1. Do not touch any non-sterile surface with sterile gloves

11. Continued preparation of sterile field

11.1. If performing full sterile technique, with sterile gloves, arrange all the sterile instruments on the sterile field. Use needle drivers to safely place scalpel blade onto scalpel handle (if using). If performing sterile tip technique, arrange instruments and sterile supplies without contaminating surfaces that must remain sterile (using a sterile instrument to arrange the supplies is helpful).

11.1.1. If sterile tip technique is planned for additional surgeries using the same instruments,

arrange the sterile field so that the “sterile tip” zone is well demarcated from the “contaminated handle” zone on the sterile field.

11.1.2. Ensure the sterile gauze, cotton tipped applicators, suture, etc is in the “sterile tip” zone of the sterile field.

11.2. Check animal’s surgical plane of anesthesia (breathing rate, color, toe pinch withdrawal reflex (can use sterile forceps or gauze to protect sterile fingers – do not place contaminated objects back into sterile field)). Adjust anesthetic depth if required.

12. Surgery

12.1. Drape animal with sterile drape with fenestration (hole) over the planned surgical incision

12.1.1. Do not contaminate surfaces that should remain sterile.

12.2. Perform required surgical procedure

12.2.1. Keep blood loss to a minimum by gentle tissue handling and hemostasis

12.2.2. Keep all exposed tissues moist with sterile saline or LRS

12.2.3. Prevent contamination of surgical site and sterile field

12.2.4. Monitor surgical plane of anesthesia as describe above at least every 10 minutes and adjust as required

12.2.5. Monitor and maintain physiological status (heart rate, respiratory rate and pattern, color of extremities and temperature and provide necessary supportive care as described in SOP ACC-2012-01 – Rodent Anesthesia throughout surgical procedure and during recovery from anesthetic

12.3. Once surgical procedure is completed, suture muscle layers and skin separately using appropriate suture material and suture pattern (see References).

12.4. Gently remove any blood from the surgery site or surrounding fur.

12.5. Ensure corneas are lubricated – apply additional lubrication if they appear dry.

12.6. Provide additional SQ fluids (10 ml/kg) if surgery was longer than expected, urine production is high (such as when using xylazine or dexmedetomidine) or if more blood loss than expected.

13. Recovery of animal

13.1. Turn off gas anesthesia (if using) and continue to provide oxygen and heat support until animal begins to move.

13.2. Move animal to warmed recovery cage and continue to provide heat support.

13.2.1. Do not recover directly on loose bedding as the bedding can enter the eyes or mouth. Place animal in empty cage with paper towel covering the bottom.

13.2.2. Monitor closely until animal begins to move around on its own and begins to groom face.

13.3. Place food and supplemental water source (i.e. Transgel, hydrogel) on cage bottom so that animal can easily access it.

13.4. Provide “pre-made” nest for post-surgical mice so they can maintain their own body temperature.

13.5. Complete surgical log with description of surgical procedure, any complications or expected outcomes and all analgesics, anesthetics, and drugs given.

13.6. Place cage flag card on cage to indicate what procedure animal had performed and all drugs administered.

13.7. Begin appropriate monitoring sheet and record when sutures are planned to be removed.

13.8. Return animal to housing room when able to move about cage normally and maintain its own body temperature

13.9. Monitor according to approved animal care protocol until animal healed and doing well and complete required monitoring sheets. Pay particular attention to surgical incision, sutures, weight and signs of pain.

13.9.1. Record all additional analgesics and supportive care on daily monitoring sheet.

13.10. External skin sutures should be removed once incision healed – typically 7-14 days.

13.11. If animal removes sutures and requires resuturing or if incision is infected, underlying tissues are exposed or the incision fails to heal normally, contact UBC Clinical Veterinarians for consultation.

13.12. Re-suturing an opened incision should only occur with Veterinary consultation and approval and only if incision is clean, shows no discharge, and is resutured within the same working day.

14. Additional surgeries using sterile tip technique

14.1. Blood should be cleaned off the instruments with water and the tips placed into a hot bead sterilizer for 15 seconds ((typically about ½ the length of the instrument can be inserted into the hot bead sterilizer)

14.1.1. Once removed from the hot bead sterilizer, the sterile “tips” that will be in contact with the tissues must be placed into the “sterile tip” area of the field while the contaminated handles must be in the “contaminated handle” area of the field.

14.1.2. Care must be taken to place and re-place the instruments properly so as to not contaminate the sterile area of the field.

14.1.3. The instruments will be very hot once removed from the hot bead sterilizer so should be given at least 20 minutes to cool to room temperature.

14.1.3.1. It is helpful to let the instruments cool while prepping the next animal for surgery.

14.2. Anesthetize and prep the animal as previously described.

14.3. Add additional sterile supplies (suture, drapes, etc.) to the sterile area of the field.

14.4. Wash hands and arms as previously described but don non-sterile exam gloves

14.5. Drape animal while handling only the underside of the drape so the upper surface remains sterile

14.6. Ensure non-sterile hands and contaminated handles of the instruments do not touch any sterile surface including the top surface of the drape, suture material, etc.

14.7. Instruments can be used for up to 6 sterile tip surgeries before needing to be re-autoclaved

14.8. Recover the animal and complete surgical log and monitoring sheets as described above.