SURGERY COMMANDMENTS

1. Thou shalt carefully plan all aspects of surgical procedures in advance
2. Thou shalt ensure the animal is appropriately anesthetized
3. Thou shalt use aseptic techniques
4. Thou shalt minimize tissue trauma
5. Thou shalt minimize blood loss
6. Thou shalt keep the patient warm
7. Thou shalt ensure personnel have been sufficiently trained for all procedures
8. Thou shalt provide post-operative monitoring and supportive care
9. Thou shalt monitor and track outcomes; taking corrective action as necessary
10. Thou shalt smile under your mask, even though no one can see you
**Objectives:**

1. General ideas to consider when planning survival surgery
2. Teach how to assess an animal’s surgical plane of anesthesia
3. Teach methods for instrument, patient and surgeon preparation
4. Perform surgery using aseptic techniques (from incision to wound closure)
5. Teach proper post-surgical monitoring and recording

**Instruments:**

All instruments must be properly sterilized. Ensure the instruments are cleaned and free of all organic material before sterilizing. A sterility indicator must be placed inside or on the surgical pack to confirm proper sterilization. Acceptable methods include:

1. Autoclave
2. Gas sterilization with ethylene oxide
3. Cold sterilization (see the list below of FDA approved products)
   - Instruments should not be used on more than one rodent without re-sterilization. A new sterile pack should be prepared for each additional animal. Alternately, instruments may be re-sterilized by using a hot bead sterilizer or flash autoclaving between rodents.

**Surgical Area:**

1. The surgical area should be isolated from active areas in the laboratory, doorways and ventilation supply ducts.
2. The area should be clean and uncluttered.
3. Surface areas should be easily sanitizable.
4. An animal preparation and recovery area, separate from the surgical area, should be provided. If a separate preparation area is not possible due to space constraints, cover the surgical area with a towel or drape and discard this after the animal has been prepared.

5. A heat source should be available anytime an animal is anesthetized (from induction to recovery) as rodents rapidly lose body heat under anesthesia.

6. Space near the surgery area should be available and contain sufficient lighting and room for hand scrubbing and donning of sterile gloves (where applicable).

7. Cardboard and paper products should not be stored directly above the surgery area. Sealable, plastic containers may be used for storage.

8. Chairs located in animal use areas should have an impervious, cleanable surface. No cloth chairs are allowed in areas animals will be used. (Cloth chairs should be covered with disposable plastic if they must be used when animals are present in the lab.)

9. The designated surgical area (e.g., bench top, chairs, equipment) should be cleaned and disinfected prior to and after surgery with a hard surface disinfectant. Always follow manufacturer's instructions.

Animal Preparation:

1. Apply a veterinary or pharmaceutical grade, bland ophthalmic lubricant to the eyes, since the blink reflex is lost during anesthesia. For extended procedures, reapplication of the ophthalmic lubricant should occur as needed in order to keep the eyes from drying out.
2. The area around the surgical site must be devoid of hair, since hair around the surgical site can act as a wick for bacterial infection. Hair removal may be achieved by either using a #40 clipper blade or a depilatory. In anesthetized mice, plucking to remove hair is relatively easy.

3. Prepare the surgical site by using alternating applications of skin antiseptics such as ethyl or isopropyl alcohol, followed by an Iodophor solution or Chlorhexidine scrub (Solution = 10%, scrub = 7.5%). Please see Skin Antiseptic table at the end of this Standard for more details.

4. Using sterile cotton tipped applicators or gauze, start with the first alcohol application. Starting in the center of the incision site, spiral outward in concentric circles toward the margins of the prepared area (never go back and forth over a cleansed area with the same cotton tipped applicator or gauze).

5. Follow the alcohol application with the first Iodophor or Chlorhexadine application repeating the concentric circle pattern.

6. Repeat the alcohol and Iodophor or Chlorhexididine step for a total of three times each. Use a new sterile cotton tipped applicator or gauze for each application.

7. When using Chlorhexidine as the skin antiseptic, ensure complete removal with sterile saline or water from the site before incision or prior to skin closure. Chlorhexidine can be irritating to skin.

8. Be careful not to excessively wet the animal as this can exacerbate hypothermia.
**Surgeon Preparation:**

1. Surgical personnel should wear a clean lab coat, mask, bouffant cap, and sterile gloves.
2. If performing multiple surgeries, new sterile gloves should be donned between animals.
3. When wearing sterile gloves, hands should be scrubbed with an antimicrobial soap prior to donning gloves if able. This is an added precaution to reduce the risk of post-operative infection, if the gloves tear during surgery.

**‘No Touch/Tips only’ Procedure:**

Some (micro)surgery may not require the use of sterile gloves. (Some examples are blastocyst transfer, some stereotaxic procedures, and many mouse surgeries.) To determine if your (micro) surgery requires the use of sterile gloves and/or drapes, please contact The Office of Animal Care and Use (OACU) at 966-5569 for further information.

If sterile gloves are not used, a “No Touch/Tips only” technique **must be used.** (This restricts the surgeon to using only the sterile working tips of the surgical instruments to manipulate the surgical field. The gloved hand must never touch the working end/tip of the instruments, the suture, suture needle, or any part of the surgical field. Sutures, catheters, and other sterile materials to be used in the surgery **must only** be handled with the instrument tips. Tissues must only be touched with instrument tips. Surgical personnel approved to utilize the ‘No Touch/Tips Only’ technique should wear a clean lab coat, mask, bouffant cap and gloves.)
**Placement of Drape Material:**

1. The use of a drape is recommended to prevent contamination of the disinfected surgical site. This is especially true for procedures that require exteriorization of the viscera.
2. Positioning of the drape over the surgical area should proceed with sterile gloves or instruments in order to maintain sterility.

**Equipment Manipulation:**

During some rodent surgeries, there may be a need to manipulate certain types of equipment (microscopes, anesthetic machines, drills, etc.). Such equipment should be disinfected before surgery. If sterile gloves touch objects outside of the sterile field, they are no longer sterile. Once surgery commences, adjustments and handling of equipment outside of the sterile field must be made using a piece of sterilized gauze, aluminum foil or commercially available sterile sleeve.

**Surgical Closure:**

The abdominal muscle/peritoneal layer and the skin must be closed separately. Appropriate suture material for each layer should be used. For closure of surgical incisions on the ventral surface (i.e., "underneath"), an interrupted suture pattern should be used in the muscle layer. When using sutures to close skin incisions, a monofilament material should be used (braided sutures used in skin tend to promote wound infection). An interrupted suture pattern should also be used to close the skin.
Wound clips or surgical staples may be used in the skin. However, clips or staples should not be used for closing skin on the ventral surface, since they may become contaminated with bedding. If clips, staples, or non-absorbable sutures are used to close the skin, they should be removed seven to fourteen days after surgery. Commercially available tissue adhesive products for skin closure work well on small skin incisions which would normally require one or two clips or sutures.

**Suture Selection:**

<table>
<thead>
<tr>
<th>SUTURE</th>
<th>CHARACTERISTICS</th>
</tr>
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<tbody>
<tr>
<td>Vicryl, Dexon</td>
<td>Absorbable – 60-90 days. Suture tissues</td>
</tr>
<tr>
<td>PDS or Maxon</td>
<td>Absorbable – 6 months. Suture tissues</td>
</tr>
<tr>
<td>Prolene</td>
<td>Non-absorbable - Inert</td>
</tr>
<tr>
<td>Nylon</td>
<td>Non-absorbable – Inert. General Closure</td>
</tr>
<tr>
<td>Silk</td>
<td>Non-absorbable – Tissue reactive. May wick microorganisms into wound. Easy to use and knot. NOT ACCEPTABLE FOR SKIN SUTURE.</td>
</tr>
<tr>
<td>Chromic Gut</td>
<td>Absorbable – Versatile. Causes mild inflammation, yet rapidly absorbed. NOT ACCEPTABLE FOR SKIN SUTURE</td>
</tr>
<tr>
<td>Staples and wound clips</td>
<td>Non-absorbable – Requires special instrument for removal.</td>
</tr>
</tbody>
</table>

- **Suture Gauge Selection:** Use smallest gauge suture material that will perform adequately.
- **Cutting and Reverse-Cutting Needles:** Provides edge to cut through dense, difficult to penetrate tissue (e.g. skin)
- **Non-cutting, taper point or round needles:** No edges for cutting. Used for easily torn tissue (e.g. peritoneum, intestine).
**Documentation:**
The USDA and PHS policies require proper documentation of animal care and use to assess compliance with research protocols and clinical care procedures. Dates of all observations, treatments and procedures must be recorded. Dates and times (including AM/PM) of all time-sensitive observations or treatments (post-operative evaluations, pain medication) must be recorded. Extent of records vary based on the nature of the procedure. However, at a minimum, records of the procedure must consist of animal/cage/group ID, date of procedure, type of procedure, anesthetics/analgesics used (dose, route, and time), anesthesia chart (verification of toe pinch), drugs given (dose, time), general procedures (e.g., intubation, beginning and end of surgery, etc.). See [https://research.unc.edu/files/2017/02/UNC-IACUC-Rodent-Anesthesia-Analgesia-Procedure-Record.pdf](https://research.unc.edu/files/2017/02/UNC-IACUC-Rodent-Anesthesia-Analgesia-Procedure-Record.pdf)

Any deviations from the approved protocol due to emergency need must be documented, explained, and reported to the OACU. **All records must be available for review at any time by IACUC and external regulatory officials.**

**Post-operative Monitoring and Supportive Care:**

1. Ensure that recovery occurs in a clean cage lined with a paper towel (rodents under the influence of anesthesia can aspirate corncob bedding).
2. Keep the patient warm until ambulatory.
3. Return to DCM facility when animal is in **sternal recumbence and ambulatory** in the cage
4. Complete and place the pink colored [Post-operative monitoring/analgesia card](https://research.unc.edu/files/2017/02/UNC-IACUC-Rodent-Anesthesia-Analgesia-Procedure-Record.pdf) (found in DCM animal housing rooms) on the cage. Cards can be removed once monitoring and/or analgesia administration as stated in approved application has been performed.
5. Provide all analgesics and fluid therapy as approved in your animal application.
6. Monitor all animals for any visible signs of pain (e.g. hunched posture, ruffled fur, lethargy, resentment to being handled, and decreased appetite).

7. Monitor animals for any signs of infection (e.g. swelling and redness around incision site, subtle change in behavior)

   NOTE: Some anesthetics (e.g: Xylazine, Medetomidine, and barbituates) may be reversed by administration of an anesthetic/sedative antagonist (e.g. Yohimbine, Naloxone, or Atipamezole).

Factors influencing possibility of Infection:
1. Foreign materials (e.g. non-absorbable suture, implants)
2. Increased surgery time
3. Use of incorrect suture (e.g. silk and cotton suture may act as a wick for bacteria into incision site)
4. Skin damage at surgical site (e.g. razor, dull clippers)
5. Not irrigating wound with sterile saline or antibiotic solution (this decreases the number of bacteria as well as removes blood clots and necrotic tissue)
6. Patient health (e.g. malnutrition, obesity, diabetes)
7. Tissue damage (e.g. improper instrument handling, heavy use of electro-cautery)
Relevant Links:
Aseptic Technique for Rodent Survival Surgeries
DCM Veterinarian Recommended Formularies and Regimens
Identification of Pain and Distress in Laboratory Animals
Pain Relief in Animals
Post-Procedure Monitoring and Analgesia Card
Isoflurane Vaporizer Monitoring Log Template
Isoflurane Drop Method Log Template
Anesthesia/Analgesia Drug Log Template

Sterilants and Disinfectants
FDA-Cleared Sterilants and High Level Disinfectants

Skin Antiseptics Table:

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<thead>
<tr>
<th>Name</th>
<th>Examples</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Iodophors</td>
<td>Betadine®, Prepodyne®, Wescodyne®</td>
<td>Reduced activity in presence of organic matter. Wide range of microbe killing action. Contact time required 10 minutes or read the label.</td>
</tr>
<tr>
<td>Chlorhexidine</td>
<td>Nolvasan®, Hibiclens®</td>
<td>Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. This is an antiseptic/antimicrobial skin cleanser. Contact time required, 15 minutes, or read the label. <a href="http://www.hibiclens.com/">http://www.hibiclens.com/</a></td>
</tr>
</tbody>
</table>

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

Disinfectants & Sterilants Table:
<table>
<thead>
<tr>
<th>Name</th>
<th>Examples *</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alcohols</td>
<td>70% ethyl alcohol, 70%-99% isopropyl alcohol</td>
<td>Remove gross contamination before using. Flammable. Contact time required 15-20 minutes.</td>
</tr>
<tr>
<td>Quaternary Ammonium</td>
<td>Cetylcid(®)</td>
<td>Rapidly inactivated by organic matter. Compounds may support growth of gram-negative bacteria. Contact time required 5-10 minutes or read the label.</td>
</tr>
<tr>
<td>Chlorine</td>
<td>Sodium hypochlorite (Clorox (®) 10% solution),</td>
<td>Presence of organic matter reduces activity. Chlorine dioxide must be fresh (&lt;14 Days old). Contact time required 10-15 minutes or read the label.</td>
</tr>
<tr>
<td></td>
<td>Chlorine dioxide [Clidox(®), Alcide(®)]</td>
<td></td>
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<tr>
<td>Aldehydes</td>
<td>Glutaraldehyde [Cidex(®), Cide Wipes(®)]</td>
<td>Toxic. OSHA has set exposure limits. Rapidly disinfects surfaces. Contact time required 5-10 minutes or read the label.</td>
</tr>
<tr>
<td>Phenolics</td>
<td>Lysol(®), TBQ(®)</td>
<td>Less affected by organic material than other disinfectants. Contact time required 10 minutes or read the label.</td>
</tr>
<tr>
<td>Chlorhexidine</td>
<td>Nolvasan(®), Hibiclens(®)</td>
<td>Rapidly bactericidal and persistent. Effective against many viruses. Contact time required 10-15 minutes or read the label.</td>
</tr>
</tbody>
</table>

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

Spor-Klenz is a hard surface antimicrobial product:  

Vimoba 128 is a hard surface disinfectant product:  

Virkon is a hard surface disinfectant product:  

For any further assistance or information, please contact an Office of Animal Care and Use Training and Compliance Coordinator at 966-5569.
GLOSSARY:

**Antimicrobial:** “destroy or inhibit the growth of microorganisms”

**Antiseptic:** “preventing the growth of microorganisms”

**Asepsis:** “state of being free of pathogenic organisms”

**Bactericide:** “destroy bacteria”

**Contamination:** “process of infecting by contact or association”

**Disinfect:** “to free from infection especially by destroying harmful microorganisms”

**Pathogen:** “a specific agent (bacterium or virus) of disease.”

**Sanitize:** “to make more acceptable by removing unpleasant or undesired features”

**Disinfectant:** “a chemical that destroys vegetative forms of harmful microorganisms (as bacteria and fungi) especially on inanimate objects”

**Sterilant:** “to free from living microorganisms”

**Sterile:** “free from living organisms and especially microorganisms”

**Sterilization:** “to make sterile”
Supply and Vendor Information

UNC-CH Hospital Pharmacy sells 100ml and 250ml bottles of isoflurane. All grant orders need to be submitted through online link below. Once ordered, it is reviewed by the pharmacy manager. If approved, orders are processed and shipped by the Shared Services Center in Durham.

https://www.uncmedicalcenter.org/uncmc/patients-visitors/amenities/pharmacies/shared-services-center-pharmacy/

Braintree Scientific
Phone: 781-917-9526
Website: www.braintreessci.com
Description: Instruments, lab equipment, isothermal pads, tattoo paste

Fisher Scientific
Phone: 800-766-7000
Website: www.fishersci.com
Description: Lab equipment, chemicals, instruments, pharmaceuticals

Henry Schein
Phone: 800-872-4346
Website: www.henryscheinvet.com
Description: Veterinary supplies, instruments, pharmaceuticals
*Need Vet License or Researcher DEA license*

Kent Scientific
Phone: 888-572-8887
Website: http://www.kentscientific.com/
Description: Surgical equipment, telemetry equipment

Med-Vet International
Phone: 800-544-752
Website: www.shopmedvet.com
Description: Veterinary supplies and instruments (discounted)
*Need Vet License*

National Band and Tag
Phone: 859-261-2035 Website: www.nationalband.com
Description: ID tags, ear tags

Patterson Veterinary
Phone: 800-225-7911
Website: www.pattersonvet.com
Veterinary supplies, instruments, pharmaceuticals including Pentobarbital
*Need Vet License or Researcher DEA license*

Plas Labs
Phone: 800-866-7527
Website: www.plas-labs.com
Description: Rodent restrainers, equipment
Roadrunner Compounding Pharmacy  
Phone: 1-877-518-4589  
Contact (as of 2/17): Coy Privette  
CoyP@Roadrunnerpharmacy.com

Roboz  
Phone: 800-424-2984  
Website: www.roboz.com  
Description: Specialize in instruments

UNC-CH Materials Management and Distribution  
Phone: 966-5671  
Description: Scientific Storeroom, General Storeroom, Chemical Storeroom

Vetamac  
Veterinary anesthesia service and manufacturer  
Cynthia A. Burns (as of 2/17), North Carolina  
Service Technician/Sales Associate  
cynthia.burns@vetamac.com

Southern Anesthesia Surgical Inc.  
Phone: 800-624-5926  
https://www.sasrx.com/  
This is a human source company that has a Veterinary division, will set up an account without a vet license
The University of North Carolina at Chapel Hill  
IACUC Training Record  
Aseptic Technique Certification Sheet

<table>
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<th>Instructor Initials</th>
<th>Technique</th>
<th>Comments</th>
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<tr>
<td>✅</td>
<td></td>
<td>Aseptic Technique:</td>
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<td></td>
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<td>- Instrument preparation/sanitization</td>
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<td>Wound Clip/Staple Application &amp; Removal</td>
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Instructor Signature: __________________________

Proficiency Rating:  
I  
II  
III

Please add email address if you are a Lab Animal Coordinator: ________________________________

Phone #: ________________________________

I certify that I have received the above training:

Student Print Name: ________________________________  
PID: ________________________________

Student Signature: ________________________________  
PI: ________________________________

Date: ________________