GUIDELINES: Aseptic Technique for USDA-covered Species Survival Surgeries

The following Guidelines are provided to ensure correct aseptic technique for all USDA-covered species survival surgeries.

**Surgical Location Approval**

DLAM maintains an area in Berryhill Hall that is approved by the IACUC for survival surgery in USDA covered species. If the procedures are not planned to be performed in this DLAM operated facility, then the location must first be evaluated and approved by the Office of Animal Care and Use (OACU) Staff. This evaluation occurs independently of the IACUC protocol review process and must be specifically scheduled by the Principal Investigator through contacting the OACU staff at 919-966-5569.

* Surgical Area

1. The surgical area should be a dedicated space or facility approved and regularly inspected by the IACUC for this purpose.
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. A heat source should be available for recovery if an extended wake-up time is anticipated. A heat source should be available during an extended surgery, since animals rapidly lose body heat under anesthesia.
5. The surgery area should contain sufficient lighting.
6. Do not store cardboard and paper products at the surgery site. Sealable, plastic containers may be used for storage.
7. Chairs located in animal use areas should have an impervious, cleanable surface. No cloth chairs are allowed in areas animals will be used.

* Surgical Preparation

1. Animal Preparation (Consult DLAM veterinarians for appropriate materials for use in non-mammalian species.)
2. Apply a 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.
3. 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 clipper blade or a depilatory.
4. Prepare the surgical site by using ethyl alcohol or isopropyl alcohol, followed by an Iodophor solution or Chlorhexidine scrub (Chlorhexidine is known to be irritating to skin).
5. Using gauze, start with the first alcohol application. Follow the alcohol application with the first Iodophor or Chlorhexidine 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 gauze).
6. Repeat the alcohol and Iodophor or Chlorhexidine step for a total of three times each.
7. When using Chlorhexidine as the skin disinfectant, ensure complete removal with sterile saline or water from the site before incision or prior to skin closure.
8. Use a new gauze for each application.

**Surgeon Preparation**

Surgical personnel should wear a mask, bouffant cap, sterile gloves and, depending upon the procedure, a sterile gown.

**Placement of Drape Material**

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. Positioning of the drape over the surgical area should proceed with sterile gloves or instruments in order to maintain sterility. Sterile surgical towels may be draped and clamped around the surgical site in place of (or in addition to) a sterile drape.

**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

Instruments should not be used on more than one animal without resterilization. A new sterile pack should be prepared for each additional animal.

**Equipment Manipulation**

During some 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 you are wearing sterile gloves to maintain asepsis and touch objects outside of the sterile field; your gloves 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.

**MONITORING THE ANESTHETIZED PATIENT**

“Careful surgical monitoring and timely attention to problems increase the likelihood of a successful surgical outcome” (Guide). Careful surgical monitoring includes **confirmation of anesthetic depth**, **maintenance of anesthesia**, and **monitoring of vital signs**.

**Confirmation of Anesthesia Depth**

The animal must be maintained on a plane of anesthesia appropriate to the surgical intervention from immediately before a surgical procedure begins until the procedure is finished. This includes the time after post-operative analgesics have been administered before they are
expected to be effective. For most species, the following techniques can be used to ascertain that the animal is appropriately anesthetized.

- **Toe pinch.** Brief clamping of the web of skin between toes or claws with a hemostat or fingers. Firmly pinching multiple toes should not elicit a withdrawal response from an animal at a surgical depth of anesthesia.

- **Palpebral reflex.** Gently tapping the medial canthus of the animal’s eye should not elicit a blink or eye flutter. This technique is not always reliable in all animals (e.g. swine).

- **Jaw tone.** The animal’s jaw should remain slack when gently extending the mandible. If the jaw is “tight” and clenched, then the animal’s anesthesia depth may not be deep enough for surgery.

- **Corneal reflex.** Touching the edge of the cornea with a gauge sponge or cotton swab will produce a good reflex if the patient is too light on anesthesia. Movement of the eyelids is an indication that the depth of anesthesia is not sufficient to do surgery.

**Maintenance of Anesthesia**

Anesthetized animals should not be left alone. Each animal responds differently when under anesthesia, therefore it may be necessary to modify your use of anesthetics during the procedure. All routinely used anesthesia options must be described in the IACUC protocol as well as a plan of how and by whom anesthetic emergencies will be addressed. Anesthetists must be highly skilled in not only delivering the anesthetic to the patient, but also in identifying anesthetic related problems.

**Monitoring Vital Signs**

The anesthetist must continuously monitor the animal patient’s basic physiological function for the duration of the procedure—from induction through recovery. At a minimum, the following vital signs are recommended to be monitored with documentation every 5-10 minutes (see DLAM Anesthesia Sheet for a suggested template):

- **Respiratory Rate (RR)** can be assessed by watching the rising and falling of the chest, by monitoring end-tidal CO2, or by artificial ventilation.
- **Heart rate (HR)** may be monitored manually (stethoscope) or with ECG or Doppler.
- **Body temperature** should be measured using a thermometer or rectal probe or other device
  - Hypothermia often occurs due to anesthesia-induced vasodilation and from surgery via opened body cavities.
  - During any surgical procedure, the animal’s body temperature should be maintained by a heat lamp, a [covered] recirculating water heating pad, forced-air warming (Bair-Hugger) or by covering the animal with warm drapes/towels.
  - DLAM Veterinarians may assist in choosing an appropriate method for your species.

For best practices, it is recommended that additional monitoring techniques be employed depending on the procedure; such as pulse oximetry, blood pressure, ECG, arterial blood:gas parameters, and end-tidal CO2 when appropriate for the animal model and procedure.
**Surgical Closure**

The fascial layer and the skin must be closed separately. Appropriate suture material and suture pattern for each layer should be used. When using sutures to close skin incisions, a monofilament material is preferred (braided sutures used in skin tend to promote wound infection).

Wound clips or surgical staples may be used in the skin. If clips, staples, or nonabsorbable sutures are used to close the skin, they should typically 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.

**ANESTHETIC AND POSTOPERATIVE RECOVERY**

**Anesthetic Recovery**

“Particular attention should be given to thermoregulation, cardiovascular and respiratory function, and postoperative pain or discomfort during recovery from anesthesia” (Guide). The anesthetic recovery period may last from minutes to hours.

- Animals should be placed into a clean recovery area in sternal or lateral recumbency. Sternal recumbency is preferred.
- As during the procedure, temperature, respiratory rate and heart rate should be monitored, evaluated, and documented during the recovery period.
- Animals should NOT be left unattended until they have completely recovered from anesthesia.

**Postoperative Recovery**

- During the post-surgical period, animals must be appropriately monitored for signs of pain and/or distress. In most species, signs of pain include decreased activity, abnormal posture, increased attention to surgical site, and gait abnormalities.
- The cardinal signs of infection include heat, swelling, redness, pain, and/or exudation. Consult a DLAM Veterinarian for any abnormal medical condition.
- The frequency and length of observation may depend on the degree of invasiveness of the procedure and the individual animal. A written plan of observation must be outlined in the IACUC protocol for each procedure proposed. The laboratory staff should observe animals at least daily following major procedures.
- If the health of the animal is questionable, a DLAM veterinarian should be notified for treatment recommendations, or to help assist in determining clinical endpoints for the animal (i.e. euthanasia).

**RECORDKEEPING FOR USDA‐SPECIES**

- The USDA and PHS require proper documentation of animal care and use to assess compliance with research protocols and clinical care procedures. All entries must be legible, made in ink, and initialed by the individual making the entry.
- 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.
- Any deletions from the record must consist of making a single line through the entry and

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initialing and dating next to the line.

- Clinical records should be kept in the vicinity of the animals and/or records must be readily accessible to the DLAM veterinary services staff and authorized inspectors.
- The USDA requires that all records be maintained for a 3 year period after the final disposition of the animal. Extent of records vary based on the nature of the procedure; however, at a minimum, records of the procedure must consist of: Animal ID, date of procedure, type of procedure, anesthetics/analgesics used (dose, route, time), anesthesia chart (vital signs – e.g. pulse rate, heart rate), drugs given (dose and time), general procedures (e.g. intubation, beginning and end of surgery, extubation, etc.). Any deviations from the procedure as approved in the protocol due to emergency need must be documented and explained.

If you need further information or if you need to review aseptic techniques, please visit [http://research.unc.edu/iacuc/](http://research.unc.edu/iacuc/), call the Office of Animal Care and Use at 966-5569 or consult DLAM veterinary services, 843-3407.

**Sterilants and Disinfectants**

**Hard Surface Disinfectants** (the designated surgical area should be cleaned and disinfected prior to and after surgery; e.g., bench top, chairs, equipment): Always follow manufacturer's instructions.

A disinfectant is a germicidal chemical substance that kills microorganisms on inanimate objects, such as instruments and other equipment that cannot be exposed to heat.

**Skin Antiseptics**

An antiseptic is a chemical agent that either kills pathogenic microorganisms or inhibits their growth. The term antiseptic is reserved for agents applied to the body.

**Instrument Sterilants** (each surgery requires the use of sterilized instruments). Always follow manufacturer's instructions.

Sterilization is the complete elimination of microbial viability, including both the vegetative and spore forms of bacteria.

**For a full list of common and approved disinfectants, please see the rodent Aseptic Surgery Guidelines.**