SMALL ANIMAL (e.g. RODENTS AND BIRDS) SURVIVAL SURGERY

STANDARD OPERATING PROCEDURE

In order to provide small animals with the highest care during survival surgical procedures, an aseptic technique is required.

Preparation of the Procedural Site

- The procedural site must be dedicated to that purpose while surgery is conducted. This area should be separated from high-traffic areas and free of unrelated equipment and supplies. If possible, the procedural site should be subdivided into separate areas for animal preparation, surgery, and recovery.
- The surgery table and associated equipment (e.g., stereotaxic apparatus) must be sanitized prior to use. Suitable products for disinfecting the surgery area include a disinfecting soap and water rinse, 70% alcohol, and quaternary ammonium based disinfectants.
- Covering of the surgical surface with clean paper (e.g., plastic-backed lab bench paper) or cloth will help prevent hypothermia and absorb fluids.

Preparation of the Surgical Instruments and Supplies

- Surgical instruments and implantable materials must be sterilized by an approved method. Sterilization may be achieved via autoclave or by other means as approved by the IACUC. Autoclaving generally is the preferred method for sterilization because of its convenience, and efficacy. Depending upon the nature of the surgical procedure, the degree of instrument contamination during the surgery, and the type of animal, a sterile set of instruments may be used for up to 2-5 small animals during the same surgery session. Instruments must be decontaminated during the procedure and between animals using a point heat source, such as a glass bead sterilizer. Exercise care to allow the instrument to cool down before use.
- Instruments and other autoclavable supplies such as gauze pads and drape material can be easily placed within disposable sterilization pouches designed for that purpose or double-wrapped in reusable cloth towels/drapes. Autoclave confirmation tape or color-change indicators should be used in/on each pack that is autoclaved and the date on which the items are autoclaved should be written on the pack. Autoclaved instruments are considered sterile for variable lengths of time depending on the manner in which they are wrapped. Double cloth wrapped instruments stored in an enclosed cabinet or container and protected from moisture are recognized as sterile for up to 6 months unless the integrity of the wrapping is compromised. Scalpel blades should be purchased sterile and not autoclaved as they are dulled by autoclaving.
- The same methods used to sterilize instruments may be applicable to implantable materials. Some materials may be commercially available as a sterile product (e.g. polyethylene tubing).
• Surgical instruments should be cleaned with an instrument cleaner or soap and water, rinsed, and dried after each surgery session. A soft toothbrush or test tube brush is often useful for thorough cleaning of delicate instruments. Instruments should be stored such that the cutting edges, tips, and points are protected from damage. Instruments should be sharpened periodically as needed.

Preparation of the Animal

• Food and water are not usually withheld from rodents unless there is concern about ingesta within the gastrointestinal tract as may occur for abdominal surgery. If this is the case, it needs to be justified in the approved protocol. Food and water withholding is not recommended in rodents for more than 12 hours.

• Preparation of the animal is usually best done in an area close to, but separate from, the surgery area.

• Plucking or shaving with electric clippers are preferred techniques for removal of fur/hair. Depilatories and razor shaving should be used carefully due to the potential for dermal irritation. Loose hair can be removed with a vacuum, tape, or wet gauze.

• The depilated area should extend well beyond the surgical margins so as to facilitate the maintenance of aseptic technique during surgery, but not so far as to contribute to hypothermia.

• Gauze sponges and cotton-tipped applicators are convenient means to wash, rinse, and disinfect the surgical site. The use of cotton tipped applicators are preferred because they help to minimize loss of body heat that may be associated with excessively moistening the skin surface.

• The surgery site must be aseptically prepped including removal of hair and disinfection by an approved method. For rats and guinea pigs washing with iodine or a chlorhexidine-based surgical soap (e.g. Betadine scrub or Nolvasan scrub) followed by disinfecting with 70 percent alcohol; alternating with an iodine solution is acceptable. For mice, 3 applications of 70 percent alcohol alternating with 3 applications of iodine solution can be used.
  o A new gauze sponge or cotton-tipped applicator should be used for each application.

Surgical Technique

• Sterile gloves, a surgical mask, cap, and a clean outer garment (e.g., lab coat or scrub top—not street clothes) are required. Exam gloves may be used provided that they are disinfected with a product such as Spor-Klenz (or other IACUC approved disinfectant) and allowed to dry for a minimum of one minute before use.

• The surgery should be conducted so as to minimize trauma to the tissues and preserve the sterility of the instruments and the surgical field. It should be completed as quickly as possible without compromising technique; tissues should be handled delicately and depending on the nature of the surgery, kept moist with warm, sterile saline. Sutures and staples should not be placed too tightly. A subcuticular skin suture pattern will often preclude the chewing and removal of sutures by the animal.
Whenever possible, the surgery site should be draped with sterile cloth, paper, surgical gauze, or clear adhesive vinyl to minimize the risk of contamination to the surgery site. Care should be taken to avoid placing a drape such that the animal cannot be monitored. Drapes can have the added benefit of keeping the animal warm.

Animals should be kept warm using an external heat source, particularly for procedures of any significant length (i.e., longer than 30 minutes). An electric or water circulating (preferred) heating pad, an overhead heat source such as a lamp or hand warmers can be used. Great care must be taken to prevent overheating or burning the animal. In all cases the external heat source should be separated from the animal by a towel or other protective barrier.

Anesthetic agents must be suitably scavenged to prevent inadvertent exposure to humans or animals. A F/Air canister or fume hood can be used. If F/Air canisters are used, they are weighed before and after each use. After an increase of 50 grams over the initial weight the canisters are disposed of properly. For prolonged procedures, particularly those accompanied by blood loss, warmed fluid therapy should be administered. The recommended amount is equal to 1-2 cc per 100 g body weight per hour of anesthesia plus any blood loss. Because of the small size of the patients covered by this policy, the intraperitoneal or subcutaneous routes are usually used.

During the surgery the animal’s respiration, tissue color and response to aversive stimuli should be monitored so that corrective action can be taken promptly if necessary.

**Recovery from Surgery**

- Depending upon the nature and duration of the surgery, it may be necessary to provide the post-operative patient with an external heat source during the recovery period. As described above, steps should be taken to protect the animal from the heat source. At the very least, animals should be placed on absorbent material or substrate or provided with other external insulating material (e.g. a towel). Animals recovering from anesthesia or surgery should not be recovered in a wire bottom cage.

- During the recovery period, the animal’s clinical condition should be monitored. Specific observations should include the body temperature, respiratory pattern, condition of the surgical wound, and the strength and rate of heartbeat.

- Rodents can often be stimulated to breathe in the case of apnea using gentle chest compression or inflating the lungs with a rubber bulb (from a pipette) applied to their nostrils. Supplemental oxygen may be beneficial.

- Animals must be monitored until they have recovered satisfactorily from anesthesia, i.e., normal respiration, sternal posture and moving about.

- The date and a brief description of the surgical procedure, including any drugs administered and the anesthetic agent(s) used, and analgesics used, must be noted on the animal’s cage card.

- Mice and rats cannot remain in investigator laboratories or other unapproved housing areas for longer than twenty-four hours without IACUC approval. USDA-covered species cannot remain in unapproved housing areas for > 12 hours.
Post-Procedural Monitoring

- Animal must be monitored post-surgically as often as necessary to assure their well-being, depending upon the nature of the procedure and the condition of the animal. Post-surgical monitoring may range from once daily for one or two days to multiple times per day for extended periods.
- Any abnormal findings must be recorded on the cage card and the veterinarian should be contacted directly or through the facility manager.
- Conditions of observation are reviewed by the IACUC at the time of protocol review. In some cases, such as when the same procedure is conducted on many animals, alternative methods of record keeping (other than on a cage card) can be used but should be kept near the animals for review. Please contact your facility manager to discuss alternative means of record keeping.

Responsibilities:

- Office of Research Compliance
  - Facilitate ongoing review of this procedure
  - Report deviations from the procedure to regulatory agencies and accrediting bodies
- IACUC
  - Ensure investigators are educated about this procedure
  - Review this procedure at appropriate intervals
  - Investigate concerns related to these standards
- Principal Investigators
  - Ensure all personnel involved in surgical procedures are trained on this procedure
  - Maintain surgical records and permit the IACUC access to surgical records