POLICIES AND GUIDELINES FOR RODENT SURVIVAL SURGERY

The following guidelines and policies for performing survival surgery on rodents are based on recommendations in the Guide for the Care and Use of Laboratory Animals (1). Survival surgery is defined as any surgical procedure from which the animal is allowed to recover from anesthesia. Issues of asepsis, surgical technique and training, anesthesia, monitoring and support, analgesia and postoperative care are all addressed in the Guide. At the same time, it is recognized that modifications of standard aseptic techniques may be necessary or desirable for rodent surgery. The policies and guidelines reflect such modifications while adhering to the intent of the Guide.

The following material is divided into guidelines, intended to provide useful information, and policies, denoted by bold print. At a minimum, policies must be followed whenever a research study involves survival surgery on rodents. Exceptions to these procedures for scientific reasons may be made if documented in the protocol and approved by the Animal Care Committee.

Preoperative Care

1. Health Status - Animals should be in optimal health at the time of surgery. An acclimation/quarantine period of 7 days is recommended to allow for general health evaluation and some degree of environmental adaptation. Rodents should be weighed pre-operatively in order to allow accurate dosing of drugs and to provide a base-line for post-operative recovery.

   Animals will undergo a minimum 7 day acclimation/quarantine prior to surgery.

2. Anesthesia – The depth and duration of anesthesia must be appropriate to the surgical procedure. Signs of adequate anesthetic depth in rodents include: regular breathing, pink ears and mucous membranes, minimal or absent pedal reflex - i.e., the animal does not withdraw the foot when the toes are firmly pinched - and absent or reduced palpebral reflex - i.e., no blink when the eyelid is touched. Movement in response to surgery indicates inadequate anesthesia.

   Preoperative fasting is rarely necessary. Withholding food for a short period, (1-4 hrs, not over night), before surgery may be useful to reduce the large variances in body weight caused by gastrointestinal contents but fasting for more than a few hours is simply an additional stressor. There is seldom any reason for withholding water.
Anyone administering anesthetics must be adequately trained/experienced in using the anesthetic selected in the species to be used. The correct dose, route, injection technique and method of restraint must be considered, as well as familiarity with the effects of the drug(s). If this is not the case, it is expected that the person will seek the assistance/training from the staff of Comparative Medicine.

**Animals will be appropriately anesthetized by trained, experienced personnel.**

Analgesia – While the degree of post-operative pain varies with the procedure and technical expertise of the operator, pain is an invariable sequel of surgery. Analgesics are used to minimize intra-operative and post-operative consequences of pain. Displaying visible signs of pain is not a survival trait in rodents, so that pain and distress will generally be concealed by the animal to the greatest extent possible. By the same token, when such signs are evident, it may be assumed that the pain is severe. Appropriate analgesics will be provided at the dose level, frequency and duration required to ameliorate post-operative distress. Exceptions to analgesic use must be justified by the investigator, and reviewed and approved by the Animal Care Committee.

**Appropriate analgesia will be provided perioperatively.**

3. **Surgical Site Preparation** – In most cases, the surgical site should be free of hair. The shaved or clipped area should provide a wide margin around the incision. The surgical area should be washed with a surgical scrub, which is removed with clean water. This is followed by application of a surgical antiseptic. Suggested products include povidone-iodine (Betadine®), chlorhexidine (Nolvasan® or Hibiprep®), or enhanced chloro-xylenols (Techni-Care®). If alcohol is used, it should be allowed to dry completely before an incision is made. Use care to avoid applying surgical antiseptics to the eyes. Swabs or small sponges should be used to avoid excessive wetting of the skin and fur adjacent to the surgically prepared site.

After the animal is anesthetized, hair will be removed from the surgical site – a #40 blade on an electric clipper is usually suitable.

Mice used for transgenic production need not be clipped if an antiseptic is used and care is taken to avoid carrying hair into the wound.

The surgical site will be cleaned using a surgical scrub.

A surgical antiseptic will be applied to the area with fresh gauze square, and allowed to remain as a final prep.
Surgical Care

1. **Surgeon** - Anyone performing surgery must be adequately trained/experienced in the procedures to be done. The Department of Comparative Medicine can offer advice, training or referrals for most surgical procedures on experimental animals. The surgeon's hands must be properly washed before aseptically putting on sterile surgical gloves. Anyone touching sterile surgical equipment or the surgical site must wear sterile gloves. Once gloved, the surgeon should touch only the prepared surgical site, sterile field and sterile instruments. A clean scrub top will reduce the risk of contamination, but in any case, long sleeves are likely to increase the risk of contamination and should be avoided. If working alone, the surgeon should have the animal anesthetized and positioned, and the surgical site prepared before opening the instrument pack and donning sterile gloves.

**Surgery will be performed by trained, experienced personnel.**

The surgeon will use sterile gloves and change them as necessary to avoid contamination.

2. **Surgical Facility** - This can be a room or portion of a room, remote from high traffic, which is easily sanitized and not used for other purposes during the time of surgery. The area, including operating boards, etc., should be physically cleaned before and after surgery, and disinfected before surgery. Accumulations of blood, feces, urine, hair, etc., should be removed completely.

Adequate space should be provided to permit establishment of a sterile field for instruments and for patient draping. Adequate light must be used to fully illuminate the surgical site.

**Surgery will be conducted on a clean, uncluttered bench or table. The area will be cleaned and disinfected before and after surgery.**

3. **Surgical Equipment** – surgical instruments, supplies, and drapes should be sterile. If multiple surgery

Steam autoclaving is the best means of sterilizing most surgical instruments. Items that are heat-sensitive, such as catheters, can usually be sterilized by exposure to ethylene oxide (gas sterilization). A few types of supplies may be sterilized or disinfected by immersion in a "cold" sterilant. When liquid sterilants are used, the directions must be closely followed and very thorough rinsing with sterile water or saline is necessary before use to avoid tissue damage. Surgical instruments exposed to liquid sterilants will deteriorate much more quickly than those sterilized by autoclaves. The Department of Comparative Medicine will provide advice
and assistance with instrument maintenance, packaging and sterilization methods.

All surgical instruments, drapes, sutures, catheters or any other supply, device or equipment which comes into contact with the surgical site will be sterilized by an appropriate method before use.

A suitably large sterile towel or tray should be used as a sterile field to maintain instruments and supplies during the procedure. To further limit contamination, instruments used for opening the skin and subcutaneous tissues should be kept separate from those used for deeper penetrations and procedures. Separation can be achieved with a second tray or by using a distinct area of a larger towel or instrument pad.

Multiple surgeries using the same instruments can present special problems. Under these circumstances, extra effort is needed to maintain the sterility of the instruments: animals should be clipped and prepared in a different area; care must be taken not to contaminate the instruments when a new animal is being positioned; the instruments should be wiped clean of blood and body fluids between each case, and may be further wiped with a small amount of alcohol or disinfectant kept in a sterile bowl for the purpose. If alcohol is used, it should be allowed to dry before the instrument is re-used: other disinfectants require thorough rinsing to avoid tissue damage from the toxic properties of the disinfectant. For long procedures, gloves should be changed between each procedure. For short procedures gloves should be changed every 3 – 5 cases, or when contaminated.

4. Draping - Draping is important for all major surgical procedures as well as for more minor procedures such as vessel cannulation or subcutaneous implants which involve implantation of a foreign body. Surgical drapes cover a wide area beyond the wound, in order to prevent contamination from the adjacent skin and hair, as well as from the surrounding bench and equipment. For rodent surgery, custom made cloth drapes work well, but are expensive and must be washed with each use. Paper drapes made from disposable autoclave wrappers, with a fenestration for the surgical site, are inexpensive, disposable, and easy to prepare and to sterilize. They are, however, somewhat stiff and light, and may be somewhat clumsy until familiarity is gained. Regardless of the material, a drape must be large enough to cover the animal and to provide a wide sterile working area.

Sterile drapes will be used for all but the most minor procedures.

Mice used for transgenic production need not be draped if aseptic procedures are applied to the surgical instruments, operating environment and technique.

5. Other Life Support Measures - Loss of body heat and fluids is of particular concern during rodent surgery. Good surgical technique includes keeping
exposed tissue moist with warm sterile saline. Body temperature should maintained during surgery and recovery, using an external heat source, such as a warm water circulating pad, gel-filled pads, or other suitable means. Electric heating pads can be used, but particular care is needed to avoid burns and hyperthermia. Passive measures to reduce heat loss include additional padding to reduce conductive heat loss, draping to reduce convective heat loss, warm subcutaneous fluid to return heat and compensate for evaporative losses, and good surgical technique to minimize operating and anesthesia time.

6. Rodents undergoing general anesthesia often develop exposure keratitis because their eyes are not completely closed and they cannot blink. This is easily prevented by applying a small amount of a bland ophthalmic ointment to the eyes at frequent intervals until corneal reflexes return. The ointment may be obtained from the Department of Comparative Medicine operating room or nursing staff.

An external heat source, such as a circulating warm water pad, will be used to minimize the animal's loss of body heat during surgery.

Bland ophthalmic ointment will be used to avoid eye lesions.

Postoperative Recovery

1. Recovery from anesthesia - Anesthesia is, in itself, a stressor. Some anesthetics induce prolonged hypothermia, aggravate fluid losses, and result in prolonged post-operative recovery. During the anesthesia recovery period, the animals must be kept warm, dry, clean and comfortable. Rodents should be placed on a soft, warm surface, such as a folded cloth towel, or other compliant material. Blood, urine, etc., should be removed by gentle cleaning. Direct contact with normal cage bedding is not recommended because it tends to cause eye injuries and respiratory and intestinal blockage in unconscious and semi-conscious animals.

Supplemental heat will reduce the metabolic demands on the animal and speed recovery, as will warm subcutaneous fluids. Many anesthetics impair thermoregulation for prolonged periods, even after apparent recovery, so that prolonged heat support may be needed to avoid depression and hypothermia after the animal is returned to normal housing. For certain drugs, such as xylazine or medetomidine, specific antagonists are available which will further hasten recovery. These measures are especially important for animals undergoing lengthy procedures (> 30 min) or animals in which injectable anesthetics have been used. In many cases, animals have depressed respiration during recovery so that a low flow of supplemental oxygen can be used to maintain adequate oxygenation.

Animals will be fully recovered from anesthesia before being returned to normal housing.
Rodents will be recovered from anesthesia in a warm environment. A heating pad or equivalent should be used to maintain normal body temperature.

2. Clinical Recovery - Animals must be monitored post-operatively, at least once daily for 3 to 5 days including weekends and holidays by the investigator and/or animal care personnel. For animals which have undergone major surgical procedures, the frequency and duration of observation may need to be increased. A plan for post-operative observations and care should be instituted together with a means for recording observations and treatments.

Care should be taken to ensure adequate food and water intake, to provide analgesics if post-operative pain/distress is evident, and to clean and dress surgical sites as needed. Weights should be recorded at regular, frequent intervals until recovery is clearly evident.

Antibiotics should not be used routinely. In the event that antibiotics are indicated for medical or scientific reasons, the drug, dose, and duration should be selected in accordance with good medical practice to achieve efficacy against the bacteria of interest. Extrapolation of antibiotic choices and doses from common practice in human medicine is unwise and may be disastrous.

Animals will be monitored post-operatively, at least once a day for 3-5 days, or until recovery is complete, by the investigator or designate, and appropriate information recorded.

A written record of postoperative observations and treatments should be kept.

Antibiotics will not be given during recovery unless specified in an approved protocol or prescribed by the attending veterinarian in consultation with the investigator.


Reviewed and approved by the Saint Louis University Animal Care Committee, May 21, 2002.