

 <p>Institutional Animal Care and Use Committee Guiding Principles and Procedures</p>	<p>Effective: May 13, 2021</p> <p>Revised:</p>
<p>Title: Experimental Surgery (Surgery for Research Purposes)</p>	<p>Page 1 of 8</p>

Guiding Principle

The use of animals in research and teaching is a privilege granted to institutions, investigators, staff, and students that commit to meeting high ethical and regulatory standards. Surgery being done for research purposes should therefore meet the requirements of the *Guide*, *USDA Policy*, and this guiding principle.

Requirements

Guide for the Care and Use of Laboratory Animals, National Research Council, 8th ed., 2011, Page 115: “Successful surgical outcomes require appropriate attention to presurgical planning, personnel training, anesthesia, aseptic and surgical technique, assessment of animal well-being, appropriate use of analgesics, and animal physiologic status during all phases of a protocol involving surgery and postoperative care.”

Guide for the Care and Use of Agricultural Animals in Research and Teaching, Federation of Animal Science Societies, 3rd ed., 2010, Chapter 2: Agricultural Animal Health Care, page 10: Includes a section on Surgery which covers the areas of multiple major surgical procedures, anesthesia and analgesia, surgery personnel, surgical facilities and aseptic technique, postsurgical care and signs of pain and distress.

USDA Animal Care Resource Guide, Policy 3 Veterinary Care, March 14, 2014 “Surgery is to be performed using appropriate anesthesia in accordance with professionally accepted medical and veterinary practice.

Animal Welfare Act (AWA) in accordance with (9CFR Ch. 1), Part 2, Subpart 2, 2.31; 4 (ix) “Activities that involve surgery include appropriate provision for pre-operative and post-operative care of animals in accordance with established veterinary medical and nursing practices. All survival surgery will be performed using aseptic procedures, including surgical gloves, masks, sterile instruments, and aseptic techniques. Major operative procedures on non-rodents will be conducted only in facilities intended for that purpose which shall be operated and maintained under aseptic conditions.”

Surgical training

Surgery must be performed by qualified personnel. Individuals must be appropriately trained to ensure that good surgical technique is practiced. All surgical procedures must be conducted as described in the approved protocol. For all procedures, normal body temperature and hydration status must be maintained.

All principal investigators, students, and animal-care personnel will have completed appropriate training before assisting with an experimental animal surgery.

All principle investigators, students, and animal-care personnel will have demonstrated proficiency in the procedure to qualified staff and documented their proficiency with the IACUC office before performing surgical procedures unassisted.

Training in aseptic technique is required prior to participation in experimental survival surgery.

Qualifications for surgeons

The Academy of Surgical Research has formulated a number of guidelines for training investigators who conduct surgery on animals. These guidelines are attached in Appendix 2. All investigators conducting surgery should be familiar with these guidelines. A principal investigator with an IACUC - approved protocol must take the responsibility of assuring that any personnel performing surgical procedures on animals under that protocol are adequately trained.

Assessment of surgeon qualifications

As part of routine protocol pre-review or Post Approval Monitoring (PAM), the attending veterinarian and IACUC may review surgical practices or surgical outcomes for ongoing projects. If deficiencies are noted, the IACUC may require further training for personnel or other changes in procedures before the project can continue.

Major surgery

Major survival surgery penetrates and exposes a body cavity, produces substantial impairment of physical or physiologic functions, or involves extensive tissue dissection or transection.

Minor Surgery

Minor survival surgery does not expose a body cavity and causes little or no physical impairment.

Survival Surgery

Survival surgery is any surgical procedure from which the animal recovers consciousness. Aseptic technique must be used for all survival surgical procedures.

Non-survival Surgery

Non-survival surgery is defined as a surgery in which the animal is euthanized before recovery from anesthesia. Non-survival surgery procedures require similar record keeping as survival surgery.

Multiple Survival Surgery

Procedure in which an animal will be used in more than one major operative procedure from which it is allowed to recover.

Guide for the care and use of Laboratory Animals

“Multiple major surgical procedures on a single animal are acceptable only if they are (1) included in and essential components of a single research project or protocol, (2) scientifically justified by the investigator, or (3) necessary for clinical reasons. Conservation of scarce animal resources may justify the conduct of multiple major surgeries on a single animal, but the application of such a practice on a single animal used in separate protocols is discouraged and should be reviewed critically by the IACUC. When applicable, the IO must submit a request to the USDA/APHIS and receive approval in order to allow a regulated animal to undergo multiple major survival surgical procedures in separate unrelated research protocols (USDA 1985, 1997a). Justifications for allowing animals not regulated by the USDA to undergo multiple survival procedures that meet the above criteria should conform to those required for regulated species. If multiple survival surgery is approved, the IACUC should pay particular attention to animal well-being through continuing evaluation of outcomes. Cost savings alone is not an adequate reason for performing multiple major survival surgical procedures.”

Animal Welfare Act

“No animal will be used in more than one major operative procedure from which it is allowed to recover, unless: (A) Justified for scientific reasons by the principal investigator, in writing; (B) Required as routine veterinary procedure or to protect the health or well-being of the animal as determined by the attending veterinarian; or (C) In other special circumstances as determined by the [Animal Care] Administrator on an individual basis.”

Survival Surgery: Rodent (based on resources available in Sudro 207)

Most surgeries on immunocompetent mice can be done on the bench.

Surgeons must wear additional PPE during the procedure. In addition to lab coat, booties, and gloves, surgeons must wear a hair cover and surgical mask when performing survival surgeries on immunocompetent mice.

Surgeries on immunocompromised mice must be done inside the biosafety cabinet (room 207A1C).

Sterilize items needed for procedures

Preparing and using the surgical station.

Clean the bench or biosafety cabinet. Spray the entire bench or hood with lemon quat solution. Leave solution on the surface for 10 minutes before wiping clean with paper towels. These paper towels can be put into the regular trash.

If working on the bench, put down clean bench covering (green, slightly tacky backed absorbent paper).

Make sure all hoses are properly connected to the vaporizer, drop box, and nose cone. Locate the purple fill adaptor for the isoflurane bottle.

Weigh and record the weight of the charcoal filters connected to the drop box and nose cone. If the filter has accumulated 50g of weight (compare new weight to beginning weight) replace with a new filter. Weigh and record the beginning weight for the new filter.

Connect the green gas hose to the back of the vaporizer and to the front of the oxygen concentrator. Plug in the oxygen concentrator. The power cord is stored in the compartment on the top. It connects to the bottom, back, center of the concentrator.

Plug in the Germinator (glass bead sterilizer) about 15 minutes before you begin your procedures. **Items placed in the glass bead sterilizer will get very hot! Allow enough time for the instruments to cool after removing them from the sterilizer before using them on the animals.**

Prepare the surgical field – put down a heating pad and plug it in. Wipe the heating pad with a Sanicare germicidal wipe. Let air dry.

Cover the heating pad with a sterile drape just before you begin. **Do not put any non-sterile items on this sterile drape.** You may open your sterile instruments and place them on this drape when you are ready to use them. Other sterile items like sterile cotton tipped applicators, sterile gauze, etc. may be placed on this field, but other non-sterile items need to be excluded.

Next to the surgical station on a cart, assemble the items you will need for your procedures.

Set up your recovery station on the bench near the sink: other heating pad and heat lamp. Keep mice in the recovery station until they are fully ambulatory before returning them to their room.

Remove and replace the sterile drape in between each surgery, or when soiled. Place the sterile drapes in the biohazard box for disposal.

Modifications for working inside the biosafety cabinet are:

Open the sash of the hood and let it run for 10 minutes before loading anything inside.

Disinfect the work surface in the hood by spraying a heavy layer of 70% ethanol on the metal tray and letting it evaporate.

Do not overload the biosafety cabinet by placing too many objects inside the cabinet.

Do not move arms in/out of the cabinet more than necessary. This disrupts the laminar air flow and prevents the biosafety cabinet from working properly.

Make sure the sash is closed enough during your procedures so that the biosafety cabinet operates normally.

The recovery station is set up on a cart directly behind the biosafety cabinet. Keep mice in the recovery station until they are fully ambulatory before placing them back on the rack.

Survival Surgery: Cattle, sheep, swine (based on resources available at the Animal Nutrition and Physiology Center and Beef Cattle Research Center)

Pre-surgical assessment of animals

It is required that all animals have a pre-procedural assessment done before every procedure. This assessment can be recorded on the anesthetic form or recorded separately and maintained by the PI as part of the animal's record.

Pre-existing health conditions may negatively affect the success of surgical procedures. Performing pre-surgical evaluations will help ensure that the animals are not overtly ill. This must include visual inspection of the animal and assessment of behavioral status. The animal must be alert and behaving normally. Physical or behavioral abnormalities must be brought to the attention of the Attending Veterinarian.

Facilities

The appropriate location for aseptic surgery is determined by the species, nature of the procedure (minor, major or emergency) and the potential for complications. Major surgery on livestock is to be conducted in a dedicated surgical suite that is used only for aseptic surgeries and the storage of essential surgical equipment. A dedicated surgical facility or space should be located outside normal facility traffic patterns & personnel access should be restricted to essential personnel. There should be separate surgical preparation and recovery areas for the animals and scrub areas for the surgical personnel. The interior surfaces of the surgical space

should be constructed of materials that are impervious to moisture and easily cleaned. Ideally, the ventilation system for the surgical area should provide a net positive pressure with respect to the surrounding facilities.

Aseptic techniques:

Major survival surgery should be conducted in a dedicated surgical suite approved by the IACUC.

1. Pre-operative guidelines in large animals:
 - a. Perform this procedure in an area separate from where the surgery is to be conducted.
 - b. Prepare the surgical site(s) with an appropriate skin disinfectant after the animal has been positioned.
 - c. Surgeons must don surgical scrubs, surgical mask, and cap, complete a surgical scrub of the hands and don sterile gown and gloves.
 - d. A separate sterile instrument pack must be used for each animal.
 - e. Animal must be draped with sterile drapes before beginning the procedure.
2. Operative guidelines in large animals:
 - a. The animal must be maintained in a surgical plane of anesthesia throughout the procedure.
 - b. A dedicated anesthetist should monitor depth of anesthesia and maintain an anesthetic record.
 - c. Close surgical wounds using appropriate techniques and sterile materials.

Anesthesia

The anesthetic regimen for any surgical procedure must be determined in consultation with the AV and must be described in the approved research protocol. Generally, inhaled anesthetics (*e.g.*, isoflurane) are recommended for long procedures that would otherwise require multiple administrations of an injectable anesthetic (*e.g.*, ketamine/xylazine). Regardless of the choice of anesthetic, a surgical plane of anesthesia must be established prior to initiating the procedure and a consistent plane of anesthesia must be maintained for the duration of surgery until euthanasia is carried out. Anesthetic depth monitoring may vary depending upon the species and anesthetic agent used.

Pain control/relief

Federal law requires pain relief according to the “*Guide*” and the Animal Welfare Act. The “*Guide*” states that “the proper use of anesthetics and analgesics in research animals is an ethical and scientific imperative”. The AWA provides requirements for animal care, treatment and practices in experimental procedures to ensure that animal pain and distress are minimized. These requirements include adequate veterinary care with the appropriate use of anesthetics, analgesics, or tranquilizing drugs or euthanasia. Care must be taken when choosing the proper dosage and schedule for a particular species, and different analgesics are indicated for different types of pain.

Postsurgical care

Animals recovering from surgery require segregation until recovery from anesthesia, clean and sanitary recovery area, adequate space, maintenance of environmental controls, and trained personnel for observation until the animal can be safely returned to original housing.

Required record keeping

1. The pre-procedural assessment can be recorded on the anesthetic form and maintained by the PI as part of the animal's record. Pre-procedural assessment should include the general appearance and health of the animal, body condition and temperature/pulse/respiration as compared to normal values. More extensive assessment may be prudent depending on the type of procedure to be conducted, and may require veterinary consultation.
2. All animals undergoing a survival or non-survival anesthetic procedure must have an anesthetic record maintained during the procedure. The anesthetic form must include a description of the procedure, the protocol number, the animal identification number, the investigators name and the anesthetist. The minimum data include: the date and starting time of the anesthetic procedure, the animal's weight, any preanesthetic assessment, anesthetic dose and route of delivery, start/stop times of the procedure, volume of fluids given, and the pulse and respiratory rate.
3. All investigators are required to create a post procedural record on all animals. This form asks for a description of the procedure performed, investigator contact information, animal identification and any treatments that the investigator or Attending Veterinarian will give to the animal. The form is used to document what treatment was done to the animal and the frequency. Antibiotics, analgesics, fluids or other drugs given to the animal must have the dose and frequency recorded, and the person giving the treatment must initial the treatment form after each treatment. This form also documents that the animal was observed following a procedure. The animal's general condition and any specific clinical signs of problems and pain assessment must be recorded.

Appropriate facilities, equipment, and trained personnel to deal with emergencies should be available for the post-surgical care of animals at all times. Surgery should be scheduled so as to ensure availability of such personnel during the postoperative recovery period.

Minor surgical procedures that do not penetrate a body cavity or produce substantial impairment (e.g., wound suturing, peripheral-vessel cannulation, certain standard agricultural practices) may be performed under less stringent conditions if performed in accord with standard veterinary practices.

Survival surgery: Dog and cat

Dog and cat neuter and spay surgeries are conducted in the Robinson Hall surgical suite using Veterinary Technology standard operating procedures and protocols.

Development of novel surgical animal models

Development of surgical techniques new to NDSU require consultation between the AV, PI and any needed consultants/experts. This consultation must include a review of available resources (personnel, equipment, space, training). If it is decided that resources outside of NDSU are needed to complete the research, a plan to obtain those resources must be designed, preferably before funding it sought by the PI.

Non-survival surgery (all species)

According to USDA Animal Care Policy #3, “Nonsurvival surgery does not require aseptic techniques or dedicated facilities.” However, there may be instances when aseptic technique is warranted. If aseptic technique is determined to be necessary, all personnel involved in these procedures must complete the Aseptic Surgical Techniques Training course through NDSU-IACUC. All personnel should follow the procedures described in their approved animal protocol.

For non-survival procedures not requiring the use of aseptic technique, the following guidelines must be followed: the surgical site should be clipped, a clean lab coat or gown and clean gloves should be worn, and the instruments and surrounding area should be clean (non-sterile instruments and supplies are acceptable). Note that procedures conducted on animals exposed to hazardous agents or animals with potential for zoonoses may require additional personal protective equipment.

Non-survival surgeries not performed in a dedicated facility must be performed in a clean area, free of clutter, and using acceptable veterinary sanitation practices analogous to those used in a standard examination/treatment room. The area immediately surrounding where surgery is conducted must not be used for other purposes during the time of surgery. Personnel present in the area must observe strict cleanliness practices for both themselves and the animals.

Use of Drugs and Other Compounds in Non-survival Surgery

All drugs and compounds administered to animals during surgery, including terminal procedures, must be of human or veterinary pharmaceutical-grade formulations when available. If non-pharmaceutical grade drugs or compounds will be used, this must be justified in the relevant IACUC protocol and approved by the Committee.

Pharmaceutical Grade controlled substances, emergency, anesthetic, analgesic, and euthanasia drugs may not be used beyond the date of expiration, even if used only for terminal procedures.