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Considerations for Rodent Surgery

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Overview

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The *Guide for the Care and Use of Laboratory Animals*[1] dictates that rodent survival surgery be performed aseptically. Aseptic technique utilizes specific practices that minimize the contamination of the surgical site, including patient preparation, surgeon preparation, sterilization of instruments and other supplies, and the use of a clean and controlled environment. Presurgical planning, intraoperative monitoring, and postoperative care are essential for successful recovery of animals from survival surgeries.

Principles

Surgical procedures are categorized as survival or nonsurvival and major or minor. Survival surgery is a surgical procedure performed on a live, fully anesthetized animal that is expected to fully recover from both the anesthesia and the procedure. Nonsurvival or terminal surgery is a surgical procedure that concludes with the euthanization of the animal before its recovery from anesthesia. Major surgery involves the exposure or penetration of a body cavity-either the thoracic cavity or the abdominal cavity-or a surgery that will cause permanent physical or physiological impairment, such as an amputation of a limb. Minor surgery does not penetrate a body cavity or cause permanent deficiency, such as subcutaneous implantation of a transponder. All surgeries require that the animal be properly anesthetized and treated humanely.[2]

Procedure

1. Presurgical Planning

Although the guidelines do not require that rodent surgeries be performed in a dedicated surgical facility, the area used must be sanitized with an appropriate hard surface disinfectant, which should be used in accordance with the manufacturer's listed concentrations and contact times. The area should also be kept free of clutter, and not be in the direct line of the supply and exhaust ducts, as the drafts could contribute to hypothermia of the animal. Access to the room should be limited when surgical procedures are conducted. An area for surgical prep (especially for the removal of the animal's hair), and for postoperative recovery and care, should also be designated and in close proximity, if not within the surgery room. In general, if surgeries are routinely performed in a specific area, the area should not be used for other activities.

Preoperative preparations should include a physical examination of the surgical patient to identify any underlying health conditions that may interfere with the surgery. Since rodents have such a high metabolic rate, and very limited fat reserves, they should not be fasted prior to surgery. The animal's hydration status should be evaluated by a skin elasticity test. The skin above the shoulders is gently lifted. In a normally hydrated animal, the skin will quickly fall back into place, whereas in a dehydrated animal, the skin will not immediately go back to its normal position. Overall appearance, such as posture and the condition of the hair coat, should also be noted. An animal that is displaying a hunched posture, or has an unkempt hair coat, may be harboring a disease. The examination should be performed just prior to the administration of anesthesia, and any abnormal conditions should be noted on the animal's chart.[2] Lastly, considerations for the anatomy and physiology of the rat or mouse must be made when preparing them for surgery.

2. Platform

Both species have a high surface area to body volume ratio making them susceptible to hypothermia during surgical procedures, especially when surgery exposes the body cavity. Efforts to prevent hypothermia include the use of a heated surgical platform.

The platform used for rodent surgery, which is usually constructed of stainless steel or hard plastic, must be covered with an insulating material or supplemental heat source to prevent the animal's body heat from escaping during the procedure. Supplemental heat sources include water circulating heating pads, forced air-warming blankets, heat lamps, or a layer of foam padding covering the surgical platform. Surgical platforms with a built in heat source are commercially available. All platforms must be of a material that is easily disinfected and impervious to moisture.

Other methods of preventing hypothermia include the use of mass insulators, placing insulating materials between the animal and the surgical platform, and using external heat sources.[3] Mass insulators entrap air within a fiber matrix, producing "still air" that surrounds the animal. Circulating hot water blankets can be used beneath the patient. This equipment is available in various sizes from rodent to equine, and allows precise thermal support with built-in thermostats.

Chemically activated heating sources can be either onetime use or reusable. One type consists of a plastic pouch filled with a chemical solution and a metal disc, which when pressed creates an exothermal reaction. This causes the liquid to solidify and release heat. Generally, they have a limited amount of heat and are only suitable for short procedures. Other chemical heating sources are available as solids at room temperature, but when heated, they become liquid. As an animal is placed on the pad, the liquid releases heat, and the pad contents solidify as they cool. These can release heat over a much longer period. As a benefit, they cannot exceed the temperature of activation (~39°C), thus eliminating the need for a thermostat.

Water packs are available as hot water bottles consisting of a rubber or silicone bag with a stopper. The packs are filled with hot water that then emits heat on the outside surface. The pack will gradually lose heat as the water cools down. A more modern version consists of a plastic sheet with water-permeable fabric adhered to the top. The space between is filled with a hydrophilic powder, which absorbs water and swells. It can be used as either a heat source or as a cooling source. Depending on the quality of the materials, it can be reused and reheated in a microwave or soaked in hot water.

Precautions must be taken when utilizing external heat sources. Body temperature should be monitored either with a rectal probe or a thermometer placed next to the animal on the heat source. All external heat sources must be checked for defects prior to use.

3. Removal of hair

The surgical site must be prepped to minimize contamination of the incision. The hair should be closely clipped or removed with a chemical depilatory cream, which dissolves the hair at the follicle.[2,3] Although the hair clipping can sometimes be performed on a conscious animal with manual restraint, the application of the depilatory cream should only be done on an anesthetized animal to prevent the ingestion of the product, eye damage, and removal of excess hair. Shaving with a razor is an option if there is no alternative. This method requires technical skill, extra time, and patience to prevent lacerations to the skin. The surgical field should be sufficiently large to allow for incision and suturing without inclusion of fur into the surgical wound, but as small as possible as to avoid the exacerbation of hypothermia.

1. Clipping
    1. Hair can be clipped using corded-electric or battery-operated clippers, preferably with a surgical A40 blade. The width of the blade should be considered. A standard 2" blade can be used for rats, whereas a ½-1" blade is more appropriate for mice.
    2. The hair is clipped against the direction of growth. Stretch the skin to stabilize it, as rodents have loose attachment of the skin to the underlying muscle.
    3. Care must be taken to avoid nicking or cutting the skin. The flat end of the blade is placed on the skin when clipping the hair. The blade should never be used with the teeth perpendicular to the skin.

Figure 1. The correct position of a hair clipper while shaving.

2. Chemical depilatory creams or lotions
    1. Apply the product to the surgical site area.
    2. After 10 minutes, the skin must be completely rinsed and cleaned of all traces of the depilatory to avoid irritation or chemical burns to the skin.

4. Surgical scrub

Surgical scrub solutions to be used should: 1) substantially reduce microbes present on the skin and contain a nonirritating antimicrobial preparation; 2) possess a broad-spectrum of antimicrobial properties; 3) be fast-acting; and 4) have persistent, cumulative activity.

The two commonly used cleansing scrub solutions are chlorhexidines and iodophors. Chlorhexidine solutions are effective against bacteria and viruses even in the presence of organic matter. In contrast, iodophors have a wide range of microbicidal action, but their efficacy is reduced in the presence of organic material; the residual activity is less than that of the chlorhexidines.

Rinses used between the scrubs are either sterile water or alcohol. Alcohol-based solutions containing 60-95% alcohol have great antimicrobial actions through the denaturing of proteins.[2] However, alcohol can be a strong skin irritant. Sterile water is effective in rinsing the area, yet it does not have any antimicrobial properties.

1. Use a moistened gauze sponge to remove gross debris including hair and dander.
2. Antiseptic cleansing solution soaked onto a gauze pad is applied to the skin starting at the incision site. Disinfection should begin along the incision line and extend outward in a circular pattern.

3. The antiseptic rinse is then applied beginning at the incision site. The skin is wiped in a circular pattern spiraling from the incision line to the periphery of the surgical field to remove the cleansing solution.
4. This is repeated three times.
5. After a final rinse, a sterile gauze pad is placed over the surgical field. This gauze may be wet with either alcohol or iodine. The gauze will be removed once the animal is transported and placed in position on a surgical platform or the surgical table/bench.

Areas of the body where the standard surgical scrubbing methods are not used include the eyes, the mouth, and the anal area. Because the surface of the eye would be damaged if scrub solutions were used, a surgical scrub is done only to the eyelids after instilling a protective ointment into the eye. In some situations, a physiologically balanced saline solution is used to flush the eye to remove gross debris and to dilute any bacteria to a level acceptable for a surgery to occur. The mouth also proves difficult to clean sufficiently for surgery. It can be rinsed with a physiologically balanced saline solution to dilute any bacteria; however, it is important to avoid using too much saline, which could cause aspiration of the fluid. Gums, teeth, and the tongue can be wiped with a nontoxic antiseptic. However, application of solutions to the mucous membranes can result in systemic absorption. Surgeries in the anal area, such as the surgical reduction of rectal prolapses, are not considered clean surgeries. The use of some antiseptic solutions can increase tissue damage and prevent or prolong healing. The use of a physiologically balanced saline solution to wash the area clean of gross debris is the preferred method of surgical preparation.[2,4]

5. Positioning

Patient positioning for abdominal procedures involves the securing of the limbs of the prone animal onto the platform with tape or a ligature. When using a ligature to extend the limbs, care must be taken to prevent circulation to the feet being compromised, to avoid excessive tension on the limbs and extreme stretching of the limbs that could impair the joints, and to avoid the impediment of breathing. The ties should be a quick release with only a half hitch loop over the limb. Some commercially available platforms come with built-in limb retraction that consists of hooks or loops of stainless steel wire or ball chain, which can be adjusted according to the size of the animal. If tape is used, it must be adhered to dry surfaces.

6. Draping

Once the animal is prepped and positioned onto the surgical platform, surgical drapes are used to prevent contamination of suture material and to maintain a sterile field at the surgical site. Drapes can be a reusable cloth material, a paper disposable material, or a disposable plastic adhesive material.

Disposable paper drapes have a woven fiber matrix for strength that allows cutting into any shape or size, including cutting a fenestration or opening in the drape, without tearing or fraying of the cut edges. They are also moisture repellent. The disposable drapes can be purchased prepackaged and presterilized in a variety of sizes and shapes. Cloth drapes are not designed to be cut by the surgeon to create a fenestration. They are purchased with a precut and bound edge fenestration. Cloth drapes require laundering and sterilization. When cared for well, cloth drapes can last for years, which makes them an economical investment.

Both paper and cloth drapes are held in place with towel clamps through the skin of the animal if it is a larger rodent, such as an adult rat. For smaller rodents, the drape is not affixed to the skin, which requires vigilance and care on the part of the surgeon to not dislodge or shift the drape once it has been positioned on the animal.

Adhesive drapes are either clear or opaque. The clear drapes are preferred for rodent surgeries, as they allow for direct visualization of the animal. Some plastic drapes are a combination of plastic and paper, with the plastic area being directly over the animal and the paper area defining the extended sterile field. The portion of the drape that is directly over the surgical incision site is designed to adhere to the incision area. The surgeon can then cut directly through the plastic when making the skin incision. Sterilized plastic wrap has been accepted as a cost-effective and useful material for rodent surgeries. Care must be taken to avoid constriction of movement for breathing when the wrap is placed around the patient. The wrap will conserve body heat, allow visualization of the patient, and provide a moisture barrier between the sterile field and the animal. It can also serve to assist in the positioning and holding of the animal for the surgery in lieu of limb fixation.

Drapes of any type should be carefully unfolded to avoid contact with nonsterile areas, equipment, and personnel; they should never be unfolded by shaking or waving.

1. Paper drapes: In the Single Drape Method, the drape is unfolded to allow for the cutting of the fenestration if one is not precut.
    1. The surgeon will place the drape over the animal keeping the hands on the side of the drape that will not touch the patient.
    2. The drape is adjusted so that the surgical field is visible through the fenestration.
    3. The drape is held in place with towel clamps through the skin of the animal in the case of larger rats.

2. Paper or plastic with an adhesive window
    1. A drape with an adhesive window requires peeling of the paper area to allow the adhesive area to stick to the surgical field.
    2. When unfolding the sterile drape, the adhesive area is generally the uppermost region and is easily accessible to the surgeon.
    3. Once the adhesive is uncovered, the drape is carefully unfolded and turned so that the sticky side is facing the animal.
    4. It is imperative that the drape is placed correctly, as once the adhesive contacts the animal it will not be able to be adjusted.
    5. The surgeon should gently press the adhesive to the surgical field to create a seal with the skin.

3. Cloth drapes
    1. Fenestrated cloth drapes are not to be cut, therefore it is the surgeon's responsibility to select a drape with a sufficiently large opening to adequately expose the surgical area, but not so large to allow exposure of any unshaved and unprepared body surfaces.
    2. The drape is carefully unfolded to reveal the fenestration.
    3. The surgeon will place the drape over the animal, keeping the hands on the side of the drape that will not touch the patient.

4. The drape is adjusted so that the surgical field is visible through the fenestration.

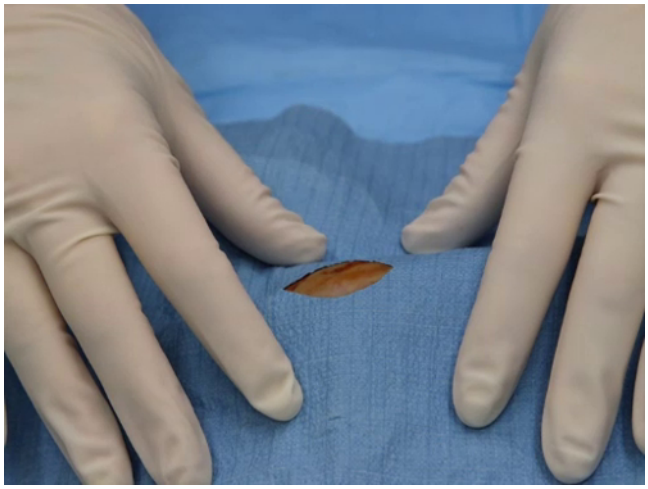

Figure 2. Surgical field visible through the fenestration of a correctly placed drape.

4. Adhesive drapes: a cost-effective and useful material for rodent surgeries.[5]
    1. The drape must be pulled from the roll in a manner to ensure the section being used remains sterile.
    2. The surgical assistant opens the box and pulls a length of the wrap out, being careful to avoid touching it to the box or any other surface.
    3. The surgeon grasps the wrap on each side, and the assistant cuts the end they are holding away (approximately 3-4 inches).
    4. After discarding the cut edge, the assistant also cuts the section from the rest of the roll.
    5. The surgeon grasps one side of the wrap and places it over the animal.
    6. The properties of the wrap allow it to adhere to all surfaces.
    7. The surgeon presses the film to the animal and creates a sterile field.
    8. There is no need to use towel clamps or worry that the drape will shift.
    9. Care must be taken to avoid constriction of movement for breathing. The wrap will conserve body heat, allow visualization of the patient, and provide a moisture barrier between the sterile field and the animal.

7. Intraoperative monitoring

Anesthetized patients must be monitored for body temperature, respirations, and heart rate until they are fully recovered.

Body temperature can be monitored directly or indirectly. For direct monitoring, a rectal probe designed specifically for rodents must be used. Small animal rectal thermometers, either mercury or digital, are too large for use in mice and rats without damage to the anal sphincter and rectal tissues. In susceptible strains, their use could precipitate rectal prolapse. Indirect monitoring involves placing a thermometer next to the animal or under the body on an external heating source. Although this will not give an exact body temperature, it can indicate the effectiveness of the heat source and allow adjustments to reduce or increase the heat as needed.

It is difficult to auscultate the heart rate and count the respirations on small rodents without specialized equipment.

Most monitoring is visual and will only indicate the presence or absence of chest or abdominal respirations. Heart rates are evaluated as present or absent by palpation or visual observation of fine movement of the chest wall. This may not be possible during a surgery due to draping and the small size of the animal.

Additional monitoring can be done through the use of electrocardiograms (ECG) and pulse oximeters. ECG's evaluate the cardiac status of a rodent during anesthesia and surgery. The pulse oximeter uses red and infrared light refraction to measure oxygen in arterial blood. This technology has been adapted for use in rodents using the tail or a paw. Both types of noninvasive ongoing measurements of the patient's vital signs are easily accessed with minimal disruption of the surgical field.

8. Postoperative monitoring

One should consider using a heating pad under the recovery cage postsurgery. In addition, pre-emptive and postoperative analgesia should be provided whenever possible. Analgesia provisions are most effective at reducing the intensity of painful stimulation when given prior to the painful event. Advantages of the pre-emptive use of analgesics include the reduction of the intensity of painful stimulation, improvement of the animal's comfort level postsurgery, the reduction in the amount of anesthesia required to maintain a surgical plane, and a smoother recovery from anesthesia once the procedure is concluded. Commonly used pre-emptive and postoperative drugs are shown in Table 1.[6]

Drug Class	Name	Dosage	Frequency
Non-steroidal anti-inflammatory drug (Noncontrolled Substance)	Ketoprofen	2-5 mg/kg SC mice 5 mg/kg SC rats	every 12–24 hours every 12–24 hours
Non-steroidal anti-inflammatory drug (Noncontrolled Substance)	Flunixin meglumine	2.5 mg/kg SC mice	every 12–24 hours
Non-steroidal anti-inflammatory drug (Noncontrolled Substance)	Meloxicam	5-10 mg/kg PO mice or 1-2 mg/kg SC mice 5-10 mg/kg PO or 1-2 mg/kg SC or PO rats	every 12–24 hours every 24 hours every 12–24 hours every 24 hours
Non-steroidal anti-inflammatory drug (Noncontrolled Substance)	Acetaminophen	50 mg/kg SC/IP or 100 mg/kg PO rats	every 8–12 hours
Opioid (Controlled Substance)	Butorphanol	0.5-3.0 mg/kg SC or 0.2-2 mg/kg IP mice 2.0 mg/kg SC 0.2-2 mg/kg IP rats	every 4 hours every 2–4 hours every 4 hours every 2–4 hours
Opioid (Controlled Substance)	Buprenorphine	0.05-2.5 mg/kg SC or IP mice 0.01 – 0.5 mg/kg SC rats	every 6–12 hours every 8–12 hours
Opioid (Controlled Substance)	Oxymorphone	0.2-0.5 mg/kg SC mice 0.2-0.5 mg/kg SC rats	every 6–12 hours every 6–12 hours

Table 1. Commonly used pre-emptive and postoperative drugs.

Summary

Through the use of aseptic technique, the incidence of postsurgical infection is greatly curtailed. Minimizing tissue trauma during the procedure, taking precautions to prevent hypothermia, controlling postoperative pain and discomfort, and the use of nutritional supplements until the animal is able to ambulate normally will all reduce the extent of negative metabolic responses to the surgical process and increase the probability of a successful survival surgery.

References

1. Institute for the Laboratory Animal Research. 2011. Guide for the care and use of laboratory animals, 8th ed. Washington (DC): National Academies Press.
2. Bernal, J., Baldwin, M., Gleason, T., Kuhlman, S., Moore. G., and Talcott, M. 2009. Guidelines for Rodent Survival Surgery. *Journal of Investigative Surgery*. 22. 445-451
3. Brauer, A., Perl, T., Uyanik , Z., English, M.J.M. ,Weyland, W., and Braun, U. 2004. Perioperative thermal insulation: minimal clinically important differences? *British Journal of Anaesthesia* 92:6. 836-840
4. Guidelines for Survival Rodent Surgery. http://oacu.od.nih.gov/ARAC/documents/Rodent_Surgery.pdf Accessed 11/11/15
5. Eakin, K., Rowe, R.K., and Lifshitz, J. 2015. Modeling Fluid Percussion Injury Relevance to Human Traumatic Brain Injury in Neurotrauma: Molecular, Neuropsychological, and Rehabilitation Aspects. Taylor & Francis Group, LLC.
6. Rodent Anesthesia and Analgesia. The University of British Columbia. Retrieved from https://animalcare.ubc.ca/conducting-your-research/rodent-anesthesia-and-analgesia