

Washington State University
Institutional Animal Care and Use Committee

Policy #6

“Aseptic and Perioperative Surgical Techniques”

Approval Date: 6/16/2022 (Replacing version: 11/24/21)

A. Purpose

To describe the requirement for aseptic technique when conducting surgery on research and teaching animals (vertebrate animals and cephalopods).

B. Background

The Animal Welfare Act and Guide for the Care and Use of Laboratory Animals require that all survival surgeries be performed using aseptic procedures. This includes the use of surgical gloves, masks, sterile instruments, and aseptic technique. Herein Principles of Aseptic and Perioperative Surgical Technique will be discussed with emphasis on the practical application of these principles in the research setting.

C. Policy

It is the principal investigator's responsibility to ensure that appropriate aseptic conditions and practices are maintained. Any surgical procedures for research, teaching or testing purposes (other than veterinary care procedures performed by a qualified veterinarian) must be included in an Animal Subjects Approval Form (ASAF) and approved by the Washington State University (WSU) Institutional Animal Care and Use Committee (IACUC) **prior** to initiating the procedure. Requests for **any variation** from the details outlined in this policy must be described within the individual ASAF for review and approval by the WSU IACUC **before the procedure can begin**. Surgery locations, surgical records and surgical support equipment and procedures may be subject to review during IACUC site visits ([Policy #34](#)).

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 non-rodent mammals 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 and 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.

A designated area such as a clean, disinfected laboratory bench is appropriate for rodent, bird, reptile, amphibian, or fish surgeries. The area should be in a non-traffic area separate from other activities during the surgical procedure. A designated area is also appropriate for minor surgery on any species.

Field surgeries for certain agricultural animals or wildlife may occur in a field, pasture or wilderness area given the proper use of aseptic technique.

Equipment:

The equipment in areas used for aseptic surgery should be easy to clean and portable to simplify sanitization of the area. The operating table or bench should be constructed with a durable surface material impervious to moisture which can be readily cleaned. Adequate lighting is essential for performing surgical procedures. Ancillary equipment such as respirators, electrosurgical units and ECG monitors should be included with the light fixtures in a routine equipment cleaning schedule.

Personnel:

Researchers conducting surgical procedures must have appropriate training. Aseptic Surgical Technique online (AST-O) and hands-on (AST-H) are required to perform survival surgical procedures as part of any WSU directed research or teaching activity. Exemptions may be requested for personnel with certain qualifications. Please refer to [Policy #20](#) for additional information on the training program and training requirements.

Sterilization of Surgical Materials:

Survival surgical procedures on all species require the use of sterile instruments and supplies. Many single-use supplies, such as needles, syringes, gloves, surgical blades, and suture materials are commercially available in sterile, ready-to-use packs. These

materials should be considered sterile until the manufacturer's expiration date, provided the storage conditions and integrity of the packaging ensure sterility of its content.

However, it is frequently necessary to (re-)sterilize (in-house) items such as surgical instruments, drapes, gauze, gowns, and catheters/devices for implant, collectively known as reusable medical devices (RMDs). Specific sterilization methods should be selected based on the physical characteristics of the materials to be sterilized. Sterilization indicators should be used to validate that all materials have achieved adequate sterilization. Please refer to the Autoclave Units section within [IACUC SOP #5: Sanitation Monitoring Practices](#) for additional information. Please refer to Appendix 1 within this document for appropriate methods of sterilization.

Re-sterilized RMDs remain sterile until an event happens (e.g., loss of integrity of the packaging (i.e., tears, punctures, broken seals, etc.), packaging becomes damp, dust accumulation, surgical pack is dropped, etc.) that compromises sterility or until the expiration date of sterilization marked externally on the packaging is reached. RMDs that become contaminated or that are past their sterilization expiration date must be properly packaged, re-sterilized, and labeled with the date of sterilization and an expiration date prior to surgical use. At the time of surgery, the surgical materials must be opened aseptically to maintain sterility.

If sterilized and stored in laboratory settings, RMDs (e.g., rodent surgery packs) must be used within 6 months of sterilization or be re-sterilized.

RMDs sterilized and stored in professionally accredited (e.g., AAHA or equivalent) medical or veterinary hospital settings under controlled temperature and humidity (range, 16-25°C [60.8-77.0°F]; 30-75%, respectively) and kept in closed cabinets or in dedicated rooms must be used within 18 months of sterilization or re-sterilized.

Because shelf life can also be affected by degradation of materials and by the amount of handling, two factors that are likely to increase over time, inventory should be managed on a "first in, first out" basis.

Non-survival surgery:

In non-survival surgery, an animal is euthanized before recovery from anesthesia. It may not be necessary to follow all the techniques outlined in this section if non-survival surgery is performed but, at a minimum, the surgical site should be clipped, the surgeon

should wear gloves, and the instruments and surrounding area should be clean. For non-survival procedures of extended duration, attention to aseptic technique may be more important to ensure stability of the model and a successful outcome.

D. Summary

The practice of aseptic technique, when performing survival surgical procedures, minimizes the chances that animal health or experimental data will be compromised by post-surgical infections. Aseptic technique requires that appropriate facilities and equipment be available and that the personnel involved be adequately trained. The key element in maintaining an aseptic environment is well-trained personnel who understand the principles of aseptic technique and utilize this knowledge on an ongoing basis.

E. References

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- CDC Guideline for Disinfection and Sterilization in Healthcare Facilities, November 2008. <https://www.cdc.gov/infectioncontrol/pdf/guidelines/disinfection-guidelines-H.pdf>
- Storage of Sterile Materials Used in Research Animals – University of Wisconsin https://www.rarc.wisc.edu/iacuc/acapac/2011-041-v_storage_of_sterile_materials_used_in_research_animals.html
- ANSI/AAMI ST79:2017 – Comprehensive guide to steam sterilization and sterility assurance in health care facilities.
- The Guide for the Care and Use of Laboratory Animals; Eighth Edition: <https://grants.nih.gov/grants/olaw/Guide-for-the-Care-and-use-of-laboratoryanimals.pdf>

Part I – Rodent, Amphibian, Fish and Small Reptiles and Birds, Survival Surgery (Group 1)

Purpose:

Aseptic surgical procedures are designed to prevent post-surgical infection due to microbial contamination of the incision and exposed tissues. Prevention of infection improves the welfare of the animal and eliminates a source of uncontrolled variation in the experimental results.

This section (Part 1) applies to aseptic surgery in small birds and reptiles, fish, amphibians, and rodents including mice, rats, chinchillas, degus, gerbils, guinea pigs, deer mice, voles, hamsters, and any other mammal belonging to the order Rodentia. Please refer to Part II for the aseptic surgery in larger mammalian species other than rodents (Group II).

Procedures:

I. Surgical Area:

- The surgical area should be separate from other laboratory activities and access restricted to essential surgical personnel during the time of surgery. (Designated surgery area)
- The area should be clean and uncluttered and work surfaces must be non-porous and sanitizable.
- Prepare the surgical area by removing all extraneous equipment or other materials.
- Clean the area with a hard surface disinfectant (see Appendix 1) and place a clean towel or drape material to cover the work surface.
- Provide supplemental heat (warm water blanket, heat pad, lamp, etc.) to prevent hypothermia in the anesthetized animal.
- Surgical areas for amphibians and fish must be clean but must also be rinsed well and free of residual disinfectants.
- Prepare your surgery area, arrange gas anesthesia mask, stereotaxic apparatus, etc. before unwrapping instruments and putting on gloves.

II. Instruments, Suture Materials, Towels, Gauze Pads and Drapes:

- All instruments that come in direct contact with the surgical site must be sterile. Refer to *Appendix 1* for recommended sterilants for surgical instruments and equipment.
- Any instruments, sutures, etc. soaked in chemical sterilants must be rinsed off with sterile water or saline (0.9% NaCl) before use.

- If performing surgery on more than one animal, begin with at least 2 sets of sterile instruments. Please view *Part II – Section VI* for guidance on multiple animal surgeries.

III. Animal Preparation:

- Animals scheduled for survival surgery must have completed the required acclimatization period (refer to [WSU IACUC Policy # 12](#) for Acclimation and Stabilization of Animals Used for Research or Teaching). Exceptions to this policy must be approved on an Animal Subjects Approval Form (ASAF).
- Evaluate prospective animals to ensure that they are apparently healthy.
- Do not withhold food in rodents before surgery unless specifically mandated by the protocol or surgical procedure. Water must NOT be withheld unless required by the protocol. Withholding food for greater than six (6) hours in rats or mice must be discussed with a veterinarian and approved by the IACUC.
- Complete animal preparation in an area distinct from the surgical area (Note: animal preparation includes anesthetic induction, hair clipping or feather plucking and initial scrub).
- Induce anesthesia and check anesthesia level
- After the animal is anesthetized, apply a sterile ophthalmic ointment to the eyes to prevent drying and corneal ulcers. (Note: Animals do not close their eyes when anesthetized and they do not blink.)
- For rodents, remove hair from the surgical site. Electric clippers with #40 or #50 blade or depilatory cream is recommended. The area to be shaved must be twice that expected for the surgical area if a larger incision is needed.
- For birds, small feathers are gently plucked around the surgical site. Avoid removing large feathers. Masking tape or stockinet material may be used to retract feathers.
- Move the animal to the surgical area.
- Place animal on a clean absorbent pad, over a heating pad (if appropriate), or in appropriate stereotaxic apparatus. Sterile saline moistened plastic drape material may be used for fish and amphibians.
- Position the animal. Do not overstretch the legs or bind them in such a way as to restrict circulation.
- For rodents and birds, put on clean or sterile gloves and scrub the shaved skin with a chlorhexidine or povidone iodine-soaked gauze/cotton. See Appendix 1 Table 3 for more information about skin disinfectants. Start from the center of the shaved site at proposed incision location and clean in concentric circles toward the edge of the shaved area. Discard the chlorhexidine or iodine-soaked

gauze and use an alcohol or saline soaked gauze to remove excess chlorhexidine or iodine in a similar fashion as above (starting from the center working towards the edge). Repeat the chlorhexidine or iodine then alcohol or saline scrub at least two more times (as described above) for a minimum of 3 total scrubs.

- Fish and amphibians have a protective mucous layer on the dermis, so alcohols, scrubs and detergents should not be used. Only sterile saline should be used on amphibians and sterile saline and/or dilute chlorhexidine solution (0.2%) may be used on fish. The skin surface should be rinsed with sterile saline or wiped with sterile saline moistened swabs/gauze.
- If possible, cover the animal with a sterile (recommended) drape with a fenestration (opening) over the proposed incision site. The drape minimizes contamination of the surgical area and surgical instruments. (To perform sterile draping, the surgeon must use sterile gloves). A moistened sterile plastic drape may be used with fish and amphibians.

IV. Surgeon:

- Wear a clean lab coat and remove all jewelry (rings, bracelets, watches) on the hands and wrists.
- Don a face mask for all surgeries. A hair bonnet or cap is recommended to prevent the surgeon's hair from contaminating the surgical area.
- Wash and scrub hands with a disinfectant soap, or surgical scrub brush, and dry with clean towels.
- Wear sterile gloves.
- Change gloves if they become contaminated.
- Anything touching the surgeon's gloves, drape or the sterile field must be sterile. If forceps are used to check the toe pinch response, the tips are considered contaminated.
- Sterile gauze or drape material may be used to manipulate non-sterile objects.

V. Recommendations for Surgical Procedures:

1. Incision(s)

- Check level of anesthesia immediately prior to starting surgery.
- Make the incision using a sharp scalpel or scissors.
- Check level of anesthesia again at intervals described in the ASAF or more often as needed.
- Control any hemorrhage through direct digital pressure, electrocautery, or with a hemostat and tying off vessels as appropriate.

- Perform the intended surgical procedure. Work carefully. Avoid unnecessary crushing of tissues. If interior tissues are to be exposed for any length of time, they must be periodically lavaged with sterile saline, or covered with a saline-soaked gauze to prevent drying.

2. Closure of Incision(s):

- Close the deeper tissue layers in one layer.
- Depending on the procedure, a simple, continuous suture pattern with a 3-0 or 4-0 (for rats) or 4-0 to 5-0 (for mice) synthetic absorbable suture may be used or a simple interrupted pattern using natural absorbable (chromic gut) may be used.
- Tighten all knots adequately. Only apply enough strength to the closure to appose tissue edges but not compress tissue.
- Close the skin as a separate layer using simple interrupted suture pattern with monofilament non-absorbable suture such as nylon. Medical grade tissue adhesive or staples or wound clips may also be used. Uncoated silk is not appropriate because of its wick function, predisposing to postoperative infections.
- Recommended suture materials for fish and amphibians are monofilament nylon, polydioxanone and polyglyconate. The use of tissue adhesives is not recommended.
- See *Appendix 1 Table 4* for more information about wound closure materials.

VI. **For Multiple Animals Receiving Surgery (Batch surgery):**

- Autoclave multiple sterile instrument packs or sterilize instruments between animals using a hot glass bead sterilizer or cold chemical sterilant. See Appendix 1 Table 2 for examples.
- Put on new sterile surgical gloves if they become contaminated by touching non-sterile surfaces or if they are soiled when starting surgery on a new animal. Because of the likelihood of contamination, gloves should be changed after every 3 animals.
- Follow all above procedures on the next animal. At any time in the surgery, if known or suspected contamination has taken place, the instrument should not be reused before re-sterilization.

I. **Postsurgical Care:**

1. Fish and amphibians

- Aquatic species should recover in a separate tank with clean water to

minimize trauma and reduce infection risk.

- Amphibians should be partially submerged with head and nares above water until fully awake and able to swim

2. Rodents and birds:

- Recover each animal in a separate cage. Conscious animals may injure an anesthetized animal. Place on a paper towel for recovery until sternal so that bedding does not restrict respiration.
- Recover the animal in a warm environment, for example in a clean cage placed over a heating pad, a circulating warm water heater, or chemical hand warmers covered with a clean towel. A warm water bottle or warmed saline bag covered with towel or a heat lamp can also be used. Avoid direct contact of the animal with heat source. Use the lowest level of heat possible to prevent accidental burns.
- In prolonged or very invasive surgeries, administer warmed, balanced electrolyte solution (such as Lactated Ringers Solution = LRS) given intraperitoneally (IP) or subcutaneously (SC). Administer 0.5-1.0 ml SC or IP to mice and 3- 5 ml SC or IP to rats. Larger rodent species may have an indwelling IV catheter placed and receive fluids (LRS) via IV drip during the procedure. Alternatively, SC fluids may be administered at a rate of 4 ml/kg for every hour of surgery. More may be needed if there was significant bleeding during surgery. Additional fluids should be given if the animal is dehydrated or not drinking.
- Monitor the color of pinnae (external ear) or footpad. If the color is too pink, this probably denotes overheating.
- Check respiration rate and depth every 10 to 15 minutes, until they have recovered their balance and can right themselves.
- Report any complications to OCV. The veterinarian must be consulted if recurring problems are not resolved.
- The animal must be monitored daily following surgery for a minimum of 3 days or as described in the ASAF, assessing such parameters as appetite, and wound healing. Administer analgesics and other drugs as stipulated in the protocol or as recommended by the veterinarian.
- Check on the animals the next morning. If they still appear lethargic, or do not appear to be eating or drinking, repeat SC or IP fluid administration and analgesics as directed by the ASAF or recommendation by the veterinarian.
- Remove non-absorbable skin closure materials 10-14 days post-surgery.

Records:

- Appropriate records of the surgical procedure, anesthesia and pre- and post-operative care must be maintained for all group I animals as either an individual or group record. All record notations must be signed/initialed and dated. A detailed description of surgical record requirements and sample surgical record forms are available on the [IACUC website](#).
- The cage of the animal(s) should be marked indicating that the animal has undergone surgery.

References

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- IACUC Guidelines, University of California at San Francisco, 2005.
- Guide for the Care and Use of Laboratory Animals, 8th Edition, 2011
- The Laboratory Xenopus sp., CRC Press, 2010
- The Laboratory Zebrafish, CRC Press, 2011
- Manual of Avian Practice, W.B. Saunders and Co, 1997

Part II –Survival Surgery for Mammalian Species Other Than Rodents (Group

II)

Purpose:

Aseptic surgical procedures are designed to prevent post-surgical infection due to microbial contamination of the incision and exposed tissues. Prevention of infection improves the welfare of the animal and eliminates a source of uncontrolled variation in the experimental results.

This portion of the policy (Part II) applies to dogs, cats, ferrets, non-human primates, swine, rabbits, wildlife, agricultural animals, and species other than those described in Part I.

Certain surgical procedures in agricultural animals or wildlife may be done under field or farm conditions as described within an IACUC approved protocol but still require appropriate aseptic technique.

Procedures:

I. Surgical Area:

- A dedicated surgical room should be located outside normal facility traffic patterns & personnel access should be restricted to essential surgical staff. A bench or counter-top within a laboratory or procedure room is not sufficient for major survival surgery for non-rodent mammals.
- There should be separate surgical preparation and recovery areas for the animals and separate scrub areas for the surgical personnel.
- The interior surfaces of the surgical room must be constructed of materials that are impervious to moisture and easily cleaned. Ideally, the ventilation system for the surgical area must provide a net positive pressure with respect to the surrounding facilities.

II. Equipment

- All instruments that come in direct contact with the surgical site must be clean and sterile. Refer to Appendix 1 Table 2 for more information.
- Any instruments, sutures, etc. soaked in chemical sterilants must be rinsed off with sterile water or 0.9% NaCl before use.
- A new set of sterilized instruments and sterile gloves must be used for each

animal if performing serial surgeries.

III. **Animal Preparation:**

- Animal scheduled for survival surgery must have completed the required acclimatization period (refer to [WSU IACUC Policy # 12](#) for Acclimation and Stabilization of Animals Used for Research or Teaching) unless otherwise indicated on the approved Animal Subjects Approval Form. Evaluate prospective animals to ensure that they are apparently healthy.
- Withhold food before surgery as appropriate for the species. Water should not be withheld unless required by the protocol.
- Perform the animal preparation in an area away from the surgical area (Note: animal preparation includes anesthetic induction, hair clipping and initial scrub).
- After the animal is anesthetized, apply a sterile ophthalmic ointment to the eyes to prevent drying and corneal ulcers.
- Remove hair from the surgical site. Electric clippers with #40 blade may be used. The area to be shaved must be twice that expected for the surgical area if a larger incision than planned may be required.
- Put on clean or sterile gloves and perform initial scrub of patient with a chlorhexidine or povidone iodine-soaked gauze/cotton. Start from the center of the shaved site (or start from where incision will be) and clean in concentric circles toward the edge of the shaved area. Discard the chlorhexidine or iodine-soaked gauze and use an alcohol-soaked gauze (70% isopropyl alcohol) to remove excess chlorhexidine or iodine in a similar fashion as above (starting from the center working towards the edge). See Appendix 1 Table 3 for more information about skin disinfectants.

IV. **Patient Surgical Scrub:**

- Move the animal to the surgical area.
- Place animal over a heating pad (if appropriate). The surgical approach will dictate actual animal position but some guidelines to consider are:
 1. The animal's respiratory function should not be compromised
 2. Limbs should not be extended beyond their normal range of motion and restraint straps should be padded as needed to prevent impaired venous return in extremities.
 3. Ruminants are frequently positioned on a slight incline, to minimize the potential for aspiration of rumen fluids.
- Personnel who perform the pre-surgical skin preparation should wear a cap,

mask and clean or sterile gloves when preparing the surgical scrub supplies and when opening pre-sterilized sponge and drape packs.

- Repeat chlorhexidine or iodine then alcohol scrub at least two more times in the surgery area (as described above) for a minimum of 3 total scrubs (3 surgical scrubs alternating with 3 alcohol scrubs).
- Drapes serve to isolate the surgical site and minimize wound contamination. Cover the animal with a sterile drape with a fenestration (opening) over the proposed incision site. Drapes should be positioned without the fabric dragging across a non-sterile surface.

V. Surgeon:

- The surgeon should wear surgical scrubs and dedicated shoes, or wear shoe covers. Head covers and face masks must cover all facial hair. Remove all rings, jewelry, and wrist watches before scrubbing. Fingernails should be trimmed short and cleaned with a disposable nail cleaner. Don a face mask for all surgeries
- Scrub sinks equipped with leg or foot-operated faucets are ideal. Regular faucets must be turned on, adjusted, and not touched again.
- The hands and forearms are washed for 30 to 60 seconds with a surgical scrub soap. Then a sterile brush is used to methodically scrub all surfaces of the hands, fingers, and forearms down to the elbows. Both arms are rinsed, and the process repeated starting with fingertips working down to the elbows. Contact times of 3 to 15 minutes and/or 5 to 20 strokes per surface are recommended. Dry the hands with a sterile towel.
- Put on sterile gown using appropriate gowning technique to maintain gown sterility.
- Put on sterile surgical gloves using appropriate gloving technique to maintain glove sterility.
- Arms and hands should always be held above the waist. Aseptic technique is maintained when the gowned and gloved surgical team only touches sterilized equipment within the sterile field.

VI. Aseptic technique without assistance:

- It is highly recommended that surgery be performed with a team of trained personnel so that the surgeon can focus on the surgery and other personnel can prepare and monitor the animal. The surgeon working alone faces logistical problems when attempting strict aseptic protocol as defined above.

- A proposed practical sequence of steps to minimize errors is presented as follows:
 1. Assemble all sterilized supplies.
 2. Change into scrubs.
 3. Set up table, heat pads and gas machines, check equipment.
 4. Weigh animal, induce anesthesia. Prepare animal by hair clip and shave, catheters placed as required. Perform initial patient skin scrub in prep area.
 5. Move animal to surgery area. Position and secure animal on the table.
 6. Connect to gas machine, connect accessory monitors. Start I.V. lines as required.
 7. Make certain that a stable anesthetic plane is attained.
 8. Put on cap and mask. Using clean or sterile gloves, prepare surgical site with scrub solutions.
 9. Open sterile instrument and prep packs that include sterile drape.
 10. Put on sterile gloves and drape patient.
 11. Remove gloves. Recheck stable anesthetic state. Open sterile glove and gown packs if not included in instrument pack.
 12. Perform surgical scrub of hands and forearms.
 13. Put on gown and gloves.
 14. Start surgery.

Records:

- Appropriate records of the surgical procedure, anesthesia, anesthesia monitoring and pre- and post- operative care must be maintained for all animals. All record notations must be signed/initialed and dated.
- Sample surgery records [forms](#) are available.
- The cage or pen of the animal(s) should be marked indicating that the animal has undergone surgery.

Resources

- Animal Welfare Act (Title 7 U.S.C. 21 31-2156) as amended by PL 99-198, December 23, 1980. Lang, C.M. *Animal Physiologic Surgery* . Springer-Verlag, New York, 1976.
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APPENDIX 1: Disinfectant, Sterilant, and Wound Closure Materials

Table 1. Recommended Hard Surface Disinfectants (e.g., tabletops, non-surgical equipment) *Note: Always follow manufacturer's instructions for dilution and expiration periods.*

AGENT	*EXAMPLES	COMMENTS
Alcohols	70% ethyl alcohol 85% isopropyl alcohol	Contact time required is 15 minutes. Contaminated surfaces take longer to disinfect. Remove gross contamination before using. Inexpensive.
Quaternary Ammonium	Roccal®, Quatricide®	Rapidly inactivated by organic matter. Compounds may support growth of gram-negative bacteria.
Chlorine	Sodium hypochlorite (Clorox® 10% solution) Chlorine	Corrosive. Presence of organic matter reduces activity. Chlorine dioxide must be fresh; kills vegetative organisms within 3 minutes of contact.
Glutaraldehydes	Glutaraldehydes (Cidex®, Cetylcode®, Cide Wipes®)	Rapidly disinfects surfaces.
Phenolics	Lysol®, TBQ®	Less affected by organic material than other disinfectants.
Chlorhexidine	Nolvasan®, Hibiclens®	Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses.
Hydrogen peroxide Peracetic acid Acetic acid	Spor Klenz	Contact time 10 minutes.
*The use of common brand names as examples does not indicate a product endorsement.		

Table 2. Approved Sterilization Procedures for Surgical Instruments & Equipment (i.e. implants and catheters) *Note: Always follow manufacturer's instructions for dilution, exposure times and expiration periods.*

AGENT	*EXAMPLES	COMMENTS
High-pressure/temperature steam sterilization	Autoclave	Effectiveness dependent upon temperature, pressure and time., see WSU SOP #5 Monitoring Sanitation Practices on selecting the appropriate monitoring system to assure sterility https://iacuc.wsu.edu/documents/2016/06/sop_5.pdf/

Dry heat	Hot Bead Sterilizer Dry Chamber	Only for re-sterilization between animals when conducting surgery on multiple animals. Instruments must be cooled before contacting tissue. <i>Only tips of instruments are sterilized with hot beads.</i>
Plasma hydrogen peroxide or gas sterilization	Vaporized hydrogen peroxide or ethylene oxide gas	Ethylene oxide. Gas is irritating to tissue and all materials require safe airing time. Significant occupational safety hazard. Appropriate sterilization indicators must be used to ensure sterility.
Cold Chemical sterilants	Chlorine dioxide (Clidox®, Alcide®) Sodium hypochlorite (Clorox® 10% solution) Glutaraldehyde (Cidex®, Cetylcide®, Metricide®) Hydrogen peroxide/Acetic acid products (Actril®, Spor-Klenz®)	Comments refer to all cold chemical sterilants Only products classified as sterilants can be used for sterilizing instruments and implants for surgery. Common disinfectants (alcohol, chlorhexidine, iodine, phenols) are not sterilants. To ensure adequate sterilization, these products must be used according to the manufacturer's recommendations for sterilization. This may require several hours of contact time. Subsequent instrument removal and handling must be done using aseptic techniques in a sterile field or the items may be recontaminated. Most cold sterilants are corrosive so will limit lifespan of instruments and are not compatible with all materials. Instruments must be clean and free of organic material prior to sterilization. Instruments must be rinsed with sterile saline or sterile water to remove the chemical sterilant before use. Chemical expiration dates must be followed.
*The use of common brand names as examples does not indicate a product endorsement.		

Table 3. Skin Disinfectants

Note: Alternating disinfectants is more effective than using a single agent. For example, an iodophor scrub can be alternated three times with 70% alcohol, followed by a final soaking with a disinfectant solution. Alcohol, by itself, is not an adequate skin disinfectant. The evaporation of alcohol can induce hypothermia in small animals.

AGENT	*EXAMPLES	COMMENTS
Iodophors	Betadine®, Prepodyne®, Wescodyne®	Reduced activity in presence of organic matter. Wide range of microbicidal action. Works best in pH 6-7.
Chlorhexidine	Nolvasan®, Hibiclens®	Presence of blood does not interfere with activity. Rapidly bactericidal and persistent. Effective against many viruses. Excellent for use on skin.
*The use of common brand names as examples does not indicate a product endorsement.		

Table 4. Wound Closure Selection

MATERIAL*	CHARACTERISTICS AND FREQUENT USES
Polyglactin 910 (Vicryl®), Polyglycolic acid (Dexon®)	Multifilament, Absorbable in 60-90 days; 25-50% loss of tensile strength in 14-21 days. Ligate or suture subcutaneous tissues where an absorbable suture is desirable. Not routinely recommended for skin closure due to high capillarity.
Polydioxanone (PDS®) or, Polyglyconate (Maxon®)	Monofilament, Absorbable in 6 months; 40% loss of tensile strength in 30-42 days. Ligate or suture tissues especially where an absorbable suture and extended wound support is desirable.
Polypropylene (Prolene®)	Non-absorbable. Inert.
Nylon (Ethilon®)	Non-absorbable. Inert. General closure.
Silk	Non-absorbable. (Caution: Tissue reactive and may wick microorganisms into the wound, <i>so silk is not recommended for skin closure</i>). Excellent handling. Preferred for cardiovascular procedures.
Stainless Steel Suture/Wound Clips/Wound Staples	Non-absorbable. Requires instrument for removal.
Cyanoacrylate (Vetbond®, Nexaband®, Tissue Mend®, Gluture®)	Medical grade skin adhesive. For non-tension bearing wounds.
*The use of common brand names as examples does not indicate a product endorsement.	
<p><u>Suture gauge selection</u>: Use the smallest gauge suture material that will perform adequately.</p> <p><u>Cutting and reverse cutting needles</u>: Provide edges that will cut through dense, difficult to penetrate tissue, such as skin.</p> <p><u>Non-cutting, taper point or round needles</u>: Have no edges to cut through tissue; used primarily for suturing easily torn tissues such as peritoneum or intestine.</p>	

APPENDIX 2: Aseptic Surgery Record Requirements

- A. Defined Groups of Animals
 - 1. Rodents/Fish/Amphibians/Reptiles/Birds
 - 2. All mammals other than rodents (e.g., cats, dogs, cow, horse, pig, goat, rabbit, dear, bear)

- B. Minimal Requirements for both Groups
 - 1. Investigator's name
 - 2. Protocol number
 - 3. Species of animal
 - 4. Animal ID
 - 5. Name of the surgeon
 - 6. Location of the surgery procedure room
 - 7. Name of the procedure being preformed
 - 8. Animal weight
 - 9. Medications given (anesthetics, analgesics, fluids)
 - a. pre, intra, post-operative
 - b. Dose/amount, route
 - 10. Surgical start and end time
 - 11. Anesthesia monitoring (assessment should be every 10-15 minutes*)
 - a. Depth of anesthesia (toe, tail pinch)
 - b. Physiological function (respiratory pattern, temperature, mucous membranes)

** The values of monitoring do not need to be written down on the surgery form, a section on the form should be checked that indicates that these assessments were performed.*
 - 12. Eye ointment was applied
 - 13. Surgical note area (used only if needed)
 - 14. Surgery records for Group 1 can be on an individual level or multiple animal sheet
 - 15. Surgery records for Group 2 must be on an individual animal record

- C. Additional Requirements for Group 2 (in addition to the minimal requirements)
 - 1. Anesthesia monitoring (assessment should be every 5-10 minutes*)
 - a. Temperature
 - b. Heart Rate or Pulse
 - c. Respiration rate

**The values of monitoring must be written down on the surgery form at the specified time intervals.*

- D. Optional anesthesia monitoring for both Group 1 & 2
 - 1. SPO2
 - 2. Blood pressure
 - 3. Capillary refill time
 - 4. End tidal CO2
 - 5. Blood gases
 - 6. Body position
 - 7. Pre-anesthetic blood values (Total protein, PCV)

8. ASA status
 9. Complications (blood loss, low PCV, arrhythmias, death)
 10. Recovery information (post-op pain level, temperature, total fluids, sedation)
- E. Intubation information (if the animal is intubated for the procedure, minimal information)
1. Trachea tube size
 2. Anesthesia system used (rebreathing, non-rebreathing)
 3. Time of extubation
- F. Post-operative monitoring for both groups (minimal required information)
1. Analgesics medications given
 - a. Name
 - b. Dose/amount
 - c. Route
 - d. Date
 - e. Time
 2. Pain Monitoring*
 - a. Active vs. Inactive
 - b. Hunched, Piloerection, Porphyrian staining
 - c. Grimace Scale
 - d. Nest building

*Pain monitoring can be achieved via many different methods. The above are examples that could be used to monitor post-operative pain. The research staff can develop a custom system approved on the ASAF.
 3. Surgical site
 - a. Inflammation and drainage
 - b. Dehiscence (suture failure or wound opening) or self-trauma
 - c. This can be a section listed as "surgical site ok" yes or no
 - d. Sutures removed if applicable
 4. General Assessment (Optional for either group)
 - a. Food and fluid consumption and hydration status
 - b. Body Weight
 5. Date released from post-operative monitoring