

**University of Cincinnati
Animal Care and Use Program**

Rodent Anesthesia and System Inspection Guideline

This guideline provides general recommendations for anesthesia and inspection of anesthesia systems of laboratory rodents at the University of Cincinnati. **See Research Administration Portal (RAP) anesthesia standard procedures for recommended species-specific routes of administration and dosages.**

Questions - contact LAMS veterinary staff at lams-veterinary@uc.edu

Anesthesia: loss of sensation resulting from pharmacologic depression of nerve function or from neurogenic dysfunction.

Considerations When Performing Anesthesia

- **Acclimation period** – allow newly arrived animals to acclimate for **at least** 48 hours prior to undergoing anesthesia.
- **Age, strain, gender, and body weight** – selection of anesthetic protocol to use.
- **Corneal drying and trauma** – for procedures lasting greater than 5 minutes or when using a facemask, an ophthalmic ointment (e.g. Puralube®) should be applied to the eyes.
- **Hypothermia and hypoxia** – low body temperature or oxygen can have significant effects on heart rate and/or blood pressure, which may result in increased morbidity or mortality.

Anesthetic Delivery Mechanisms

- **A combination of inhalation and injectable anesthetics can be used.**
- **Inhalation** – preferred for safety, anesthetic plane stability, and hypoxia prevention. **Isoflurane** is preferred for both short and long procedures since it takes effect rapidly and has an extensive history of reliable animal recovery. Two delivery systems:
 - 1) Controlled delivery gas anesthesia machine-Recommended (SomnoSuite® -low flow preferred for safety, vaporizer-high flow).
 - a. Due to the small respiratory capacity in rodents, a non-rebreathing system should be used.
 - b. **Induction**
 - Use the lowest concentration of anesthetic gases needed.
 - Induction 3-5%
 - Maintenance 1-3%.
 - Whenever possible, place the induction chamber in a certified/validated chemical or snorkel fume hood OR ducted (exhausted) or carbon-filtered biosafety cabinet.
 - Attach a charcoal waste anesthetic gas canister to the exhaust port unless using a hood. See Waste gas canister section for additional information.
 - If possible, purge induction chamber with oxygen prior to opening the chamber to retrieve the anesthetized animal. If the anesthesia machine does not have an oxygen purge valve, you can still purge some of the anesthesia by turning off the isoflurane vaporizer first and wait a few seconds, then turn off the oxygen flow meter.

- If moving animal to a nose cone, do not turn off the oxygen flow. Remove the animal from the induction chamber, move the animal to the nose cone, and turn on the vaporizer.
- 2) Drop box/Bell jar method is only for ultra-short non-surgical procedures (e.g. tail clipping) or euthanasia by anesthetic overdose.
 - a. Must done in a certified/validated chemical fume hood or snorkel fume hood to ensure removal of waste anesthetic gases, and to minimize occupational exposure to exiting gases.
 - b. The animal cannot come in direct contact with the anesthetic.
- **Injectable** – Intraperitoneal (IP) is the most common and preferred route.
 - Other routes: subcutaneous (SC), intramuscular (IM [large rodents only]), and intravenous (IV, catheter suggested).

Anesthesia monitoring and recovery

- **Anesthesia monitoring parameters**
 - Respiratory rate/pattern – observe movement of chest wall and abdomen.
 - Mucous membrane color – should be pink (oxygenated), not blue or grey (hypoxic).
 - Muscle relaxation and response to stimuli – observe loss of pedal withdrawal (toe pinch) or eye blink reflexes and lack of response to surgical stimulus (e.g. incision).
- **Reducing anesthesia depth**
 - Injectable anesthesia: use an appropriate reversal agent (refer to IACUC protocol or consult LAMS veterinary staff) and administer supplemental oxygen via face mask or nose cone.
 - Inhalation anesthesia: turn off isoflurane vaporizer (leave oxygen on) until condition improves; then turn vaporizer back on.
- **Prevent hypothermia**
 - Use a recirculating warm water blanket or isothermal heat source (e.g. Gaymar Stryker Heating/Cooling T/Pump, Snuggle®Safe) during anesthesia. Do not place the animal directly on the heat source; cover the heat source with a towel or sterile drape. Electric heating pads are discouraged since they heat unevenly and pose an increased risk of thermal injury.
 - Covering an animal with a sterile drape or gauze helps to conserve body heat.
 - Administering warm sterile 0.9% sodium chloride or lactated ringers SC or IP (5-10ml/kg/hr) may be indicated.
- **Recover animals from anesthesia** in a separate clean cage without bedding (use sterile paper towel for substrate). As anesthetized animals are unable to move away from heat sources, ensure recovering animals are placed only partially on a covered heat source to minimize overheating. Continually monitor the animal until sternal, and place into home cage when fully ambulatory. Provide food and fluid supplements on cage floor if needed.

Waste Anesthetic Gas Canister Use, Placement and Disposal

- Charcoal waste anesthetic gas (WAG) canisters must be secured in the upright position for optimal functionality.
- WAG canisters must be monitored by weight and/or hours of use.
- WAG canisters do not need to be placed below the vaporizer to be effective (JAVMA¹).

Designs and Types of Waste Anesthetic Canisters

The main active substance in WAG canisters is activated charcoal (carbon). WAG canisters can be used passively (gas flow through the canister is facilitated by the pressure of oxygen passing through the anesthesia machine vaporizer) or actively (an evacuated assist system where a motor-generated vacuum pulls gas through the canister). Active scavenging is strongly recommended for heavy use (e.g. multiple animals, daily/weekly use).

There are two designs of activated charcoal canisters:

Top vent: Reported to be more effective at removing WAG.

Bottom vent: Bottom holes must be elevated off any solid surface to work as designed.

Both vary in size from 50 gm up to 200 gm absorption capacity. WAG canisters, connected by flex tubing to the outflow of an induction chamber or non-rebreathing Bain line (animal breathing nose cone), will inherently be unstable and fall over on their side if not secured. Options pictured below.



Monitoring and Disposal of Waste Anesthetic Canisters

- 1) When a new canister is placed, a baseline weight and date must be documented on the canister.
- 2) Canisters must be weighed at a frequency that ensures they do not exceed the maximum increase allowed. For an F-air canister, a 50 gm increase typically occurs after approximately 8 hours of use.
- 3) If the canister weight exceeds manufacturer's recommended capacity (typically 50 gm more than the initial baseline weight for the smaller 50 gm canister), the canister will not perform optimally and must be replaced.
- 4) If a canister no longer gains weight or loses weight (relative humidity fluctuations can cause this), the canister should be replaced.
- 5) Anesthetic waste canisters or charcoal should be bagged (to prevent off gassing), labeled as hazardous waste with an Environmental Health & Safety (EH&S) Waste label including "charcoal" and the agent (e.g. isoflurane), and requested to be picked up by the EH&S office. **DO NOT** place in regular trash.

Anesthesia and Vaporizer Inspection Guidelines

The *Guide for the Care and Use of Laboratory Animals* (NRC 2011) notes that appropriate anesthesia is a critical component of the Animal Care and Use Program (ACUP). Properly functioning gas anesthesia machines are key to the delivery of the anesthetic agent to the animal and the safety of personnel. There are many factors to consider (e.g. frequency of use, age of machine, environmental conditions) when developing a program for anesthesia machine maintenance and upkeep, and all components need to work properly as an integrated unit.

The below guidelines are for traditional anesthesia machines with inline vaporizers and those with ventilators. Low-flow rodent anesthesia systems (i.e. SomnoSuite®) require no annual calibration.

Guidelines:

- 1) Anesthesia systems must be inspected **annually** by a qualified professional or certification company unless otherwise approved by IACUC. This inspection includes:
 - a. Full evaluation of the anesthesia machine
 - b. Efficacy test of the vaporizer
 - c. Inspection of O₂ delivery system, patient circuits, evacuation technique
- 2) If the verified delivery of the vaporizer is $\pm 15\%$ out of calibration of the target value, the vaporizer must be serviced off-site by qualified personnel or company.
- 3) The date of servicing (or the service date) must be clearly posted on the anesthesia machine (i.e., sticker affixed to vaporizer).
- 4) **Biennial Inspection Request:** A request to inspect rodent anesthesia systems every other year may be submitted as a protocol amendment to the IACUC. Acceptable justifications include:
 - a. The system is not mobile (i.e. is a tabletop system).
 - b. The system is used infrequently – a log of hours used must be kept and be made available; infrequent use must be justified in the exception request.
 - c. No modifications have been made to the system outside manufacturer recommendations.

Related Policies and Guidelines

- [EH&S Advisory 8.5 Anesthetic Gas Use \(Research\)](#)
- [Survival Surgery Guidelines](#)

References

1. American Veterinary Medical Association (AVMA), *Influence of the position of charcoal air-filtration canisters on the efficacy of waste isoflurane scavenging as assessed in a randomized experiment*, Elizabeth S. Moore, DVM¹; Erin K. Daugherty, DVM^{1,2}; Bhupinder Singh, DVM²; David E. Mooneyhan, BS²; Teresa J. Porri, PhD³ and Wendy O. Williams, DVM² *JAVMA Vol. 254, No. 12, Pages 1459-1465, June 15, 2019*, <https://doi.org/10.2460/javma.254.12.1459>