1. PURPOSE

This Standard Operating Procedure (SOP) describes methods for anesthetizing rats.

2. RESPONSIBILITY

Principal Investigators (PIs) and their research staff, veterinary care staff.

3. INTRODUCTION

3.1. Rats are not routinely fasted prior to anesthesia due to their inability to vomit.
3.2. Rodents can be anesthetized with either inhalant gas or injectable drugs. The use of inhalant gases is the preferred method of anesthesia whenever possible.
3.3. Heat loss is rapid in anesthetized rodents. Keep animals warm by covering them (e.g. gauze pad or towel) and/or providing a heat source until the animal has recovered from anesthesia.
3.4. Never leave an anesthetized animal unattended.

4. MATERIALS

4.1. Material or equipment to provide or conserve body heat (e.g. gauze pads, heating disc or pad, warm-water circulating pad)
4.2. Ophthalmic ointment (natural tears)
4.3. Gas anesthesia machine (calibrated within the last 12 months) with adequate gas scavenging system or filter
4.4. Induction chamber constructed of a see-through material (glass, polycarbonate, etc.)
4.5. Isoflurane
4.6. Ketamine (100mg/mL) *Controlled drug
4.7. Xylazine (20mg/mL)
4.8. Acepromazine (10mg/mL)
4.9. Sterile isotonic saline (0.9% saline) or sterile water for injection

5. PROCEDURES FOR ADULT RATS

5.1. Isoflurane anesthesia:
   5.1.1. Induction:
      5.1.1.1. Place the animal in the induction chamber
      5.1.1.2. Adjust the oxygen flowmeter to 0.8 to 1.5 L/min.
      5.1.1.3. Adjust the isoflurane vaporizer to 3% to 5%
   5.1.2. Maintenance:
      5.1.2.1. Use the mask connected to the Bain circuit,
      5.1.2.2. Adjust the flowmeter to 0.4 to 0.8 L/min.
      5.1.2.3. Adjust the isoflurane vaporizer to 2 to 2.5%.
      5.1.2.4. Apply ophthalmic ointment (natural tears) to both eyes to prevent dryness and damage to the cornea. Reapply as needed.
5.1.3. Recovery:
   5.1.3.1. Turn off the isoflurane vaporizer but keep the animal on oxygen.
   5.1.3.2. Transfer animal to their cage once it begins to move and allow to recover fully (sternal position).

5.2. Ketamine/Xylazine/Acepromazine anesthesia:
   5.2.1. Injectable anesthetic dose can vary with the sex, the age, the strain, and the body condition of the animal.
   5.2.2. Contact your veterinarian for advice on the appropriate dose prior to use.
   5.2.3. Anesthetic dose: ketamine 50mg/kg, xylazine 5mg/kg, acepromazine 1mg/kg.
   5.2.4. To prepare cocktail, in a sterile vial or bottle with a rubber stopper, mix:
      - 5mL of ketamine (100mg/mL)
      - 2.5mL xylazine (20mg/mL)
      - 1mL acepromazine (10mg/mL)
      - 1.5mL of sterile isotonic saline or sterile water for injection.
   5.2.5. Label as "Rodent Cocktail" and indicate expiration date on vial or bottle (maximum 6 months).
   5.2.6. Mixed cocktail should be protected from light and stored in a cool place.
   5.2.7. Administer 0.1mL/100g body weight intramuscularly or intraperitoneally.
   5.2.8. Apply ophthalmic ointment (natural tears) to both eyes to prevent dryness and damage to the cornea. Reapply as needed.
   5.2.9. Duration of anesthesia is approximately 30 minutes.
   5.2.10. After 30 minutes, a half dose may be administered as needed.

6. PROCEDURES FOR NEONATAL RATS

6.1. Hypothermia:
   6.1.1. Use only in animals less than 7 days of age.
   6.1.2. Provides immobilization and mild analgesia for short, minor procedures.
   6.1.3. Protect pup in a glove or paper-lined tube to avoid damage to the skin.
   6.1.4. Induction:
      - 6.1.4.1. Immerse pup in ice water or crushed ice for 3 to 4 minutes.
   6.1.5. Maintenance:
      - 6.1.5.1. Place pup on a paper-covered ice pack.
      - 6.1.5.2. Use a fiber optic surgical lamp if necessary as incandescent lamps will warm the animal and interfere with anesthesia.
      - 6.1.5.3. Duration of anesthesia is approximately 10 minutes.
   6.1.6. Recovery:
      - 6.1.6.1. Remove animal from ice pack and allow to warm.
      - 6.1.6.2. Recovery time can be up to 1 hour.

6.2. Isoflurane anesthesia:
   6.2.1. Neonates require higher concentration of isoflurane than adults (maintenance at 3-4%). See section 5.1 for detailed procedure.
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| 2016.03.16 | 5.2.1 and 5.2.2 (NO TEXT) | 5.2.1 Injectable anesthetic dose can vary with the sex, the age, the strain, and the body condition of the animal.  
5.2.2 Contact your veterinarian for advice on the appropriate dose prior to use. |
**Rodent Procedure Log**

**Instructions:** complete this log for rodent procedures requiring anesthesia, analgesia or post-procedure care (ex. surgeries, experimental infection). Keep the log in the housing room while active and in your files for 3 years for future review by the Quality Assistant and/or the FACC.

### ANALGESIA
- **Carprofen:** mouse: 20mg/kg, rat: 5-10 mg/kg, SC, every 24 hrs
- **Buprenorphine:** mouse: 0.1mg/kg SC or IP every 4-8 hrs; rat: 0.05mg/kg, SC or IP, every 8-12 hrs
- **Lidocaine/bupivacaine** (local analgesic)
- Other: ____________

### ANESTHESIA
- **Isoflurane** 2-2.5%
- **Ketamine/xylazine/acepromazine**:
  - Mouse: 100 mg/kg (K)- 10 mg/kg (X)- 3 mg/kg (A) IP
  - Rat: 50 mg/kg (K)- 5 mg/kg (X)- 1 mg/kg (A); IP or IM

### OTHER AGENTS ADMINISTERED
- Other: ____________

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**Comments/footnotes:**

*Dose can vary with the sex, the age, the strain, and the body condition of the animal.*

Revised: 2014-01-06
**ANALGESIA**

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- **OTHER_________________________________________
  
  Initial the appropriate boxes when completed

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