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THE HONG KONG UNIVERSITY OF SCIENCE AND TECHNOLOGY

ANIMAL & PLANT CARE FACILITY
動植物養育實驗所

# GUIDELINES ON ANESTHESIA - PART 1: MOUSE

ANIMAL AND PLANT CARE FACILITY, HKUST

# GUIDELINES ON ANESTHESIA - PART 1: MOUSE

This guideline is provided for users planning mouse procedures using injectable anesthetic agents at the Animal & Plant Care Facility, The Hong Kong University of Science & Technology (APCF-HKUST). All anesthetic agents used in the procedure must be listed on an approved protocol. Any exception must be described and justified in the user protocol, and must be approved during the regular review process prior to starting any procedure. All procedures must be performed only by appropriately trained personnel.

**Related Guidelines:**

Guidelines on Rat Anesthesia, Guidelines on Rabbit Anesthesia, Analgesia Guidelines, Antibiotic Guidelines

## I. RECORDKEEPING FOR INJECTABLE ANESTHETICS

Following the Hong Kong Regulations on Pharmaceuticals, the dispensing and use of drugs classified as Schedule 3 (Dangerous Drugs or DD) must be properly recorded in the Dangerous Drugs Register (DDR). APCF will issue whole bottle(s) of the drug(s) requested along with the DDR book to record the dispensing and use of the DDs by the PI or their authorized representative.

Along with the required DDR, data recording until full recovery of the animal(s) that has undergone any procedure(s) is also essential.

Please see attached (Form 1A) for reference.

## II. PRE-OPERATIVE CARE

1. Perform a thorough physical examination on the mouse. Unless otherwise stated on the approved protocol, only administer anesthetics to an apparently healthy animal. No need to fast the animal prior to surgery as the mouse does not have a vomiting reflex.
2. Determine the weight of the mouse to know how much anesthetic agent is needed.
3. Draw the correct amount of anesthetic agent based on the body weight.
4. Handle the mouse using the appropriate restraint technique. Inject the drawn anesthetic intraperitoneally (IP) or intramuscularly (IM) using gauge 25-27 hypodermic needle. Use the smallest size needle that will deliver the injection smoothly as this will minimize the pain of injection while the animal is still conscious. The maximum amount of fluid that can be injected intraperitoneally is 2.0-3.0 mL at a time.
5. Place the mouse gently back into its cage after the anesthetic agent is given. Wait to effect prior to starting the procedure.
6. If stated on the approved protocol, additional analgesics and/or antibiotics can also given to the animal. Please see Guidelines on Mouse Pain Management and Guidelines on Mouse Antibiotics Use.

## III. SUPPORTIVE CARE OF ANIMALS DURING ANESTHESIA

1. For any procedures requiring anesthesia longer than 5 minutes, apply an ophthalmic ointment to both eyes to prevent desiccation. Please check with the APCF Veterinary team for recommendations regarding ophthalmic ointment.
2. Maintain normal body temperature and avoid hypothermia by using thermal pads or heat lamps during anesthesia. Care must be taken when using an over-the-counter electric heating pads and lamps as these can be prone to overheating.
3. Whenever necessary, provide fluids either IP or subcutaneously (SC) to the mouse during prolonged anesthesia to maintain adequate hydration as described in the approved protocol.

## IV. MONITORING AND ASSESSMENT OF ANESTHESIA

1. Monitor respiratory rate and effort, and the color of mucous membranes at regular intervals (approximately 5 minutes each).
2. Assess level of anesthesia by pedal reflex or whole body parts movement. Adjust anesthetic delivery to maintain surgical plane by giving half dosage or less of the anesthetic agent to effect whenever necessary.

## V. POST-OPERATIVE CARE

1. Place the mouse in a warm, clean, dry, and quiet environment away from other mice. Cover or replace bedding material with autoclaved tissue (Kim Wipes®) while waiting for the animal to regain full consciousness. Bedding can stick to eyes or be inhaled while animals are recovering from anesthesia, which can lead to corneal damage and/or aspiration.
2. Provide warmth during recovery. Surgical heating pads or a 50-75 watt incandescent lamp can be used for this purpose. When using a lamp, place it 12-14 inches away and in a position that the rodent can move away from the light and heat source whenever it desires. Attentive monitoring must be done when using over-the-counter heating pads and incandescent lamps as they can lead to overheating.
3. The mouse must be continuously monitored until it can maintain an upright posture and walk normally inside the recovery cage before leaving or transferring back to its original cage.

## VI. ANESTHETIC AGENTS

The table below contains information on the injectable anesthetic agents available in APCF. Please note that this does not include other agents that may be listed in a user's approved protocol.

| Agent | Dose Rate | Effect | Duration of Anesthesia |
|---|---|---|---|
| Alphaxolone | 10 mg/kg IV | Surgical anesthesia | 5 minutes |
| Ketamine + Medetomidine | 75mg/kg + 0.5 mg/kg IP | Surgical anesthesia | 20-30 minutes |
| Ketamine + Xylazine<br>(cocktail) | 80-100 mg/kg + 10 mg/kg IP<br>(Given 0.1mL per 20.0g BW IP) | Surgical anesthesia | 20-30 minutes |
| Ketamine + Xylazine + Acepromazine<br>(administered separately) | 80-100 mg/kg + 10 mg/kg + 3 mg/kg IP | Surgical anesthesia | 30-40 minutes |

All administered anesthetic drugs classified as Schedule 4 by Hong Kong SAR laws must be properly recorded in the Dangerous Drugs Register.

The facility based the above doses from the following references:

a. Laboratory Animal Anesthesia (Flecknell)
b. Plumb's Veterinary Drug Handbook (Plumb)
C. Exotic Animal Formulary (Carpenter & Marion)

## VII. TRIBROMOETHANOL (AVERTIN®)

Tribromoethanol (TBE) is no longer available as a pharmaceutical product. PIs should not use TBE as an anesthetic but if there is justification for TBE use then PIs must make their own solutions from a non-pharmaceutical grade chemical. It should also be described on the protocol with a scientific justification for using it, and should be approved by the Animal Ethics Committee.

The disadvantages of using TBE include the following:

1. It is an irritant, especially at high doses, high concentrations, or with repeated use. Adhesions can be seen in the abdominal cavity organs following IP injections.
2. It degrades in the presence of heat or light and produce nephrotoxic and hepatotoxic byproducts. Administration of degraded solutions has been associated with mortality post-surgery.
3. It can cause intestinal ileus several weeks post-injection.
4. The effects are unpredictable in mice younger than 16 days, or in animals with altered carbohydrate metabolism, such as various mouse strains used for diabetes or obesity models (db/db mice or ob/ob mice).

## VIII. PREPARATION OF KETAMINE AND XYLAZINE COCKTAIL FOR MICE

1. Prepare a sterile injection vial.
2. Verify the concentration and expiration date of your drugs prior to mixing.
3. In preparing a 10.0 mL total stock volume or anesthetic cocktail, use [100 mg/mL] ketamine and [100 mg/mL] xylazine with the following amount:
   a. 1.75 mL ketamine
   b. 1.25 mL xylazine
   c. 7.0 mL saline or sterile water for injection
4. Each vial of the cocktail will be labeled with the following:

| | |
|---|---|
| Protocol Number | |
| Principal Investigator | |
| User | |
| Form 2 Number | |
| Animal Handling Number | |
| Mouse Anesthetic Mix | |
| Dosage | |
| Expiration | |
| Prepared by | |

The expiration date for the cocktail is set at one month from the date of mixing.

## IX. ANESTHETIC MONITORING CHART FOR MICE

| Date | PI/User | Protocol Number | Strain & Mouse ID/Code | Weight | Sex | DOB |
|---|---|---|---|---|---|---|
| Procedure | | | | | | |
| Anesthetic Agent(s) Used | | | Dose (mg) | | | |
| Time of Induction | | | Time of Recovery | | | |

| Time | Heart Rate (BPM) | Respiratory Rate (RPM) | Temperature (C) | Signature |
|---|---|---|---|---|
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## REFERENCES

AALAS Learning Library. LAT Chapter 10: Common Technical Procedures. Accessed 2 January 2019.

Office of Animal Resources, Institutional Animal Care and Use Committee, University of Iowa. IACUC Guidelines on Anesthesia. Accessed 2 January 2019.

University of California San Diego IACUC. Best Practice Use of Avertin. Accessed 23 March 2019.