Guidelines for the use of Isoflurane anesthesia without a vaporizer In rodents

The use of a calibrated vaporizer provides the most consistent and safe delivery of inhalant anesthesia. If a standard vaporizer is not available, isoflurane may be suspended in paraffin/mineral oil and administered on cotton or gauze in an induction chamber and/or nose cone. Use of isoflurane without a precision vaporizer has a very small margin of safety. Animals can easily become overdosed and die if not constantly monitored.

All research staff must be fully trained prior to initiating any new procedure. LAR staff is available to assist in the training for the use of inhalant anesthetics.

Standards for an induction chamber

- **1.** The size of the induction chamber should be sufficient to contain the animal with normal posture but small enough to reduce dead space.
- **2.** A platform is required to prevent direct animal contact with the isoflurane soaked cotton/gauze. Fine wire mesh is recommended. Solid platforms must have sufficient openings to allow circulation of vaporized isoflurane, but small enough (or cover large openings with wire mesh) to keep animal from reaching isoflurane soaked cotton/gauze.
- 3. The isoflurane suspension may permanently mar the finish of plastic.
- **4.** Ensure all ports leading into the chamber are closed to contain vaporized anesthetic within the chamber.

Standards for nose cone

- 1. The size of the nose cone should be sufficient to enclose the nose and mouth of the animal.
- **2.** There must be adequate distance between the isoflurane soaked cotton and the animal to prevent direct contact (at least 2").
- **3.** Recommendations for modified nose cones include, a syringe barrel with the plunger removed (i.e. 3-5ml for mouse, 20ml for adult rats) or plastic test tube at least 2.5" in length.
- **4.** Cotton or gauze should be forced into the closed end of a nose cone leaving at least a 2" distance between the cotton and the open end of the nose cone.

Preparation of Isoflurane Suspension

1. Isoflurane is mixed with paraffin/mineral oil to a concentration of 10%-20%.

10% suspension = 1 part isoflurane + 9 parts oil

20% suspension = 2 parts isoflurane + 8 parts oil

2. Due to vaporization of the isoflurane, the concentration of the suspension will change over time. Therefore, a fresh suspension must be mixed prior to initiating procedures and every 1-2 hours thereafter.

Preparation of the induction chamber or nose cone

- **1.** Initially charge the chamber or nose cone by placing the isoflurane suspension directly to the cotton/gauze using a glass pipette.
- 2. Variables such as chamber size, suspension concentration, room temperature and time will affect the quantity of isoflurane required to achieve anesthetic levels. It is important that a user carefully consider these variables to determine the appropriate amount of isoflurane suspension required for their chamber and/or nose cone. Over time, the isoflurane vapor will dissipate from the chamber and/or nose cone requiring more suspension to be added to the cotton/gauze.

Administration of Isoflurane

1. Differences in rodent strains and ages may affect the rate of induction and maintenance of an anesthetized animal. Most animals will become unconscious in 1-2 min following exposure to the isoflurane suspension. Therefore animals must be constantly monitored to guard against overdose.

Parameters to monitor include:

- 1. Alertness
- 2. Response to stimuli
- 3. Breathing rate

Once the animal in an induction chamber reaches the set limits of the listed parameters, remove the animal from the chamber. If maintenance of the anesthetic state is required, transfer the animal to a prepared nose cone charged with the isoflurane suspension.

- **2.** Animals placed on a nose cone must be monitored continuously to ensure maintenance of the anesthetic state and guarding against overdose. Adjustment of the anesthetic level is achieved by altering the distance of the nose into or out of the end of the nose cone.
- **3.** During recovery, animals must be monitored consistently until the animal is alert and moving normally around the cage.

Safety Precautions

Use of isoflurane must follow proper safety guidelines as outlined in the product MSDS. http://butler.compassites.com/?m=product_view&id=6708021

Due to potential for user inhalation exposure, the use of isoflurane in an induction chamber and/or via nose cone must be within a hood that is directly exhausted to the outside or has carbon filtration to trap waste gas.