

 Lab Animal Facilities	STANDARD OPERATING PROCEDURE Delivery of Inhalant Anesthetics Using a Bell Jar	Quality Form SOP # 2.A.8 Revision: 01 Last Reviewed: 11/01/23
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1.0 PURPOSE:

The purpose of this procedure is to outline the proper use of a bell jar.

2.0 SCOPE:

This procedure applies to all veterinary or laboratory staff using a bell jar to deliver inhalant anesthesia to small rodents (mice, rats and hamsters).

3.0 DEFINITIONS:

3.1 Bell Jar: A closed, lidded chamber often referred to as an open drop jar used for the delivery of inhalant anesthesia to small rodents. Bell jars are used to deliver inhalant anesthesia for very short-term procedures (i.e. 30-60 seconds) or for euthanasia methods involving inhalant anesthesia. If longer duration anesthesia is needed, an alternate method of anesthesia, such as an anesthetic machine with a calibrated vaporizer or injectable anesthetics, should be used. Bell jars are not suitable for larger species that must be anesthetized with inhalants via a calibrated vaporizer.

4.0 PROCEDURES:

4.1 Materials needed:

1. Fume hood
2. Bell jar or other glass container of known volume with tightly fitting lid.
3. Mesh platform (plastic or woven wire)
4. Cotton balls or gauze squares
5. Isoflurane
6. Recovery cage with supplemental heat source

4.2 Bell jar anesthesia contains no provisions for scavenging anesthetic waste gases. Therefore, **the bell jar must be used in a fume hood** to protect personnel from inhalation exposure to anesthetic gases.

4.3 The liquid anesthetic (isoflurane) is volatilized by placing it on the cotton or gauze in the bottom of the jar. The following chart illustrates the approximate concentration of anesthetic achieved based on volume of the chamber and volume of isoflurane used.



Example of a glass drop jar with plastic platform

Concentration of Isoflurane (%)	Internal Volume of Anesthetic Chamber				
	1 L	2 L	3 L	4 L	5 L
1	0.05 mL	0.10 mL	0.15 mL	0.20 mL	0.26 mL
2	0.10	0.20	0.31	0.41	0.51
3	0.15	0.31	0.46	0.61	0.77
4	0.20	0.41	0.61	0.82	1.02
5	0.26	0.51	0.77	1.02	1.28

*Volumes in shaded boxes are in mL and indicate the volume of isoflurane to be applied to the cotton or gauze in the bell jar.

From “Anesthesia and Analgesia in Laboratory Animals,” page 32

- 4.4 Alternatively, the isoflurane can be diluted in propylene glycol before soaking the cotton or gauze. Diluting with propylene glycol reduces the isoflurane vapor in the bell jar and creates a somewhat safer and more stable anesthetic without using a calibrated vaporizer.

1. For mice: 20% v/v isoflurane in propylene glycol
2. For rats: 30% v/v isoflurane in propylene glycol
3. Example calculation: The percent concentration of a liquid reagent is based upon volume per volume

To make 500 mL of a 20% v/v isoflurane in propylene glycol:

Volume % (v/v) = [volume of solute (mL) / volume of solution (mL)] * 100

0.20 (percent as a decimal) * 500 (total solution volume) = 100 mL of isoflurane needed

500 mL (total solution volume) – 100 mL (isoflurane volume) = 400 mL (propylene glycol needed)

- 4.5 Working inside the fume hood, soak the cotton balls or gauze with the appropriate volume of isoflurane or isoflurane/propylene glycol mixture, then place inside the bell jar.
- 4.6 Cover the isoflurane soaked cotton balls or gauze with a **plastic or wire mesh platform**. This will prevent the rodent from coming into direct contact with the anesthetic agent, which can cause localized discomfort or irritation to the animal's skin.
- 4.7 Place the animal in the bell jar and close the lid tightly. The bell jar should be large enough for the animal to comfortably move around. Only one animal at a time should be placed in the bell jar.
- 4.8 Because this method of inhalant anesthesia does not utilize a calibrated vaporizer the concentration within the jar cannot be precisely controlled and lethal concentrations of isoflurane can rapidly accumulate. The animal must be closely monitored at all times while it is in the jar. If at any time the animal has difficulty breathing (respiration becomes labored, irregular, or stops) or poor mucous membrane color (pale, blue, or gray membranes), the animal should immediately be removed from the chamber.

- 4.9 Observe the animal closely for cessation of voluntary movement and recumbency. Loss of righting reflex when the jar is gently tipped and slowed (but regular) breathing is a sign that a plane of anesthesia has been reached.
- 4.10 Remove the animal from the bell jar. Check the mucous membrane color, respirations, and withdrawal reflexes. The procedure can begin only if the animal's mucous membrane color and respirations are normal and the withdrawal and palpebral reflexes are absent.
- 4.11 When using a bell jar to deliver anesthesia there is no way to maintain anesthesia once the animal has been removed from the bell jar, thus this method only provides anesthesia for very short-term procedures, such as retro-orbital blood collection, tail snipping, or subcutaneous tumor implantation. This method of anesthesia is NOT appropriate for surgical procedures.
- 4.12 When the procedure is completed, place the animal in the recovery cage, administer any required post-operative analgesics, and monitor the animal until it is fully awake.
 1. Complete the "Rodent Anesthesia &/or Surgery Report" form. Submit the completed form to:
 - a. BEB: on the door of Room 204 (VT Office)
 - b. CTRC: on the door of Room 7071 (VT Office)
 - c. JSMBS: outside the door of Room B2107 (VT Office)
 - d. Pharmacy, Hochstetter, Park Hall: at the LAF Office at each location
 - e. RIA: Desk on 3rd floor inside LAF facility
 2. Return the animal to its regular housing only after it has fully recovered from anesthesia, and identify the cage by placing a blue Post-Operative Observation Card at the front of the cage card holder.

References:

1. Kohn, DF, Wixson, SK, White, WJ, Benson, GJ (ed): Anesthesia and Analgesia in Laboratory Animals, Academic Press, 1997. Laboratory Animals, Academic Press, 1997.
2. Itah, Refael, et al. "A Replacement for Methoxyflurane (Metofane) in Open-Circuit Anaesthesia." Laboratory Animals, vol. 38, no. 3, 2004, pp. 280–285.
3. Fox, James G., et al. "Inhalant Anesthetics." Laboratory Animal Medicine, Third ed., Elsevier/Academic Press, 2015, pp. 1141–1142.

Approvals:

Name	Title	Date of Approval
Jennifer Peirick, DVM	LAF Director/Attending Veterinarian	11/01/23
Jolie McCutcheon, DVM	LAF Clinical Veterinarian	11/01/23

Change History:

Revision #	Description of Change	Effective Date
01	Converted to new SOP format. Added exact location for submission of Anesthesia/Surgery Report by facility.	11/01/23