



Johns Hopkins University

Animal Care and Use Committee

Use of Ether for Animal Anesthesia¹

PURPOSE: This policy was developed by the JHU Joint Health Safety and Environment/Animal Care and Use Committee to provide investigators with information on the safe use of ether for animal anesthesia.

BACKGROUND: Gaseous anesthetics are often used to anesthetize rodents for experimental procedures. Such agents are classified as rapid-onset, short-acting (halothane and isoflurane) or slow onset, long-acting (ether, methoxyflurane). Such agents can be administered by using anesthetic machines with calibrated vaporizers, or by exposure in a closed container by the "open-drop" method. The open-drop method while easiest to use, can often lead to over-anesthetization and possibly death when the short-acting agents are used. Conversely, ether and methoxyflurane are relatively safer agents to use by open drop because they volatilize poorly, and it takes a longer time for animals to become anesthetized, greatly increasing the margin of safety.

Diethyl ether (ether) is a flammable liquid with a characteristic "sweet" odor. Methoxyflurane (Metofane) was the agent of choice over ether because of the flammability of ether, however, Metofane is no longer available for purchase. When Metofane was removed from the market, ether was re-approved for use in laboratory animal procedures at Johns Hopkins, **subject to approval on a case-by-case basis during protocol review by the JHU Animal Care and Use Committee (ACUC).**

If ether has been approved for your use in anesthesia by the ACUC, the following procedures must be followed in order to protect yourself and others who work in the same area in which ether is being used. Because the air odor threshold concentration for ether (8 ppm) is extremely low, it is not uncommon for other personnel working in the area in which ether is being used to become concerned regarding potential hazard. The requirements for use of ether described below are designed to permit effective anesthesia at concentrations that are below its lower explosive limit and well below the Occupational Safety and Health Administration's (OSHA's) permissible exposure limits for people.¹ Adherence to these procedures also should lessen ether odor in the vicinity of its use and thus the potential for concern by others in the same facility.

¹OSHA limits are: 400 ppm as an 8-hour, time-weighted average concentration, which is 1200 mg/cubic meter. The American Conference of Industrial Hygienists recommends \leq 500 ppm for 15 min. Threshold for mild nasal irritation (but no injury) is 200 ppm.

PROCEDURE:

What is the proper manner of storing ether?

The explosive potential of ether increases with period of storage. For example, after 12 months, there is potential for formation of explosive peroxide crystals in unopened ether containers.

1. Order ether in the smallest volume needed.
2. Date the container when received, and again when opened.
3. Ether and any device used to contain it should be stored in a vented National Fire Protection Association (NFPA) flammable-material cabinet.
4. Send opened containers of ether for disposal 6 months after first opening.
5. Send unopened ether containers for disposal 12 months after receipt.
6. Contact the Office of Health Safety and Environment at 410-955-5918 to find out the proper location for disposal and for answers to other questions about safe use of ether.

Where may ether be used?

1. Whenever possible, the use of ether anesthesia should be restricted to a properly functioning, explosion-proof, chemical fume hood.
2. Ether may be used in a biosafety cabinet that is connected to the building exhaust system, but may not be used in a biosafety cabinet that is vented into the room. ***If you don't know the venting of a particular biosafety cabinet, do not use ether there.***
3. The use of ether in a biosafety cabinet should be conducted well within the confines of the cabinet, and as far back on the work surface as possible.
4. You may check with the JHU Biosafety Officer (410-955-5918) if you are unsure of the nature of a particular type of hood or cabinet.

How much ether may be used at a time?

1. All manipulations involving volumes of ether greater than 5 ml must be performed in a properly functioning, explosion-proof, chemical fume hood.
2. The total cumulative volume of ether used in a biosafety cabinet may not exceed 5 ml, regardless of the duration of the procedure.
3. Do not bring more than 10 ml of ether to any JHU central animal facility area for use in that location regardless of the type of fume hood or biosafety cabinet you will be using.

How should ether be used for anesthesia?

1. The effective concentration of ether to produce anesthesia is 1.9%. This concentration can be produced with 0.08 ml (80 microliters) per liter of volume of a container.

2. Animals may be exposed to ether presented on a cotton ball or gauze pad inside a conical tube or small chamber, but there should not be direct contact of the animal with the cotton or gauze.
3. Induction takes 5 to 10 minutes. There are mouse strain differences in sensitivity to ether: C3H>BALB/c>DBA/2>ICR>C57B16. Ether is a satisfactory anesthetic in gerbils but it is unsafe in guinea pigs.
4. See “Use of Experimental Animals at the Johns Hopkins University” (www.jhu.edu/animalcare), and/or consult a JHU veterinarian (410-955-3273) for more information about anesthetizing animals with ether.

What is the proper way to dispose of materials used to deliver ether?

1. Allow the ether to evaporate completely from the material while still in the hood or biosafety cabinet prior to disposal.
2. Dispose of the material in a red bag.

¹Revised by the JHU Joint Health Safety and Environment/Animal Care and Use Committee February 22, 2006, reviewed September 25, 2012, January 31, 2018, January 5, 2021, January 18, 2024.