1.0 Introduction
Isoflurane is a halogenated gas that is commonly used as an inhalation anesthetic in animal research. It is a clear, colorless, volatile liquid at room temperature and pressure. Exposure to second-hand anesthetic gases has been associated with several health concerns and can occur when vapors escape into the work environment during the administration of anesthesia. These guidelines will focus on the safe use of isoflurane during research with animals.

2.0 Hazards and Risks
Isoflurane is an eye and skin irritant and central nervous system toxin. Acute exposure symptoms include:
- Nausea
- Vomiting
- Headache
- Dizziness
- Drowsiness
- Skin irritation

Long-term exposure may cause chronic or adverse health effects including:
- Hypotension (low blood pressure)
- Tachycardia (increased heart rate)
- Respiratory depression
- Reduced mental performance
- Liver and kidney disease
- Possible reproductive effects (sterility, infertility, miscarriages, and birth defects)

Conditions that may increase exposure risk to waste anesthetic gases (WAG) include:
- Not ensuring a tight seal around the animal’s nose cone
- Failing to flush the induction chamber with oxygen prior to opening the chambers to transfer animals
- Performing multiple animal surgeries, requiring an extended length of time that anesthesia is delivered
- Leaks within the anesthesia machine, breathing system, hoses, and connections
- Spills of liquid anesthetics
- Charcoal canister misuse or over saturation
- Not ensuring that procedures are conducted in areas that receive adequate room ventilation
- Not ensuring that the anesthetic gas scavenging system is connected and working effectively

3.0 Exposure Monitoring
Isoflurane exposure monitoring can be requested to assess the potential of waste anesthetic gases in the general laboratory work area. Exposure monitoring is requested by contacting Environmental Health Safety Office (EHSO) by emailing indhyg@emory.edu.

The National Institute for Occupational Safety and Health (NIOSH) Recommended
Exposure Limit (REL) for isoflurane is 2.0 parts of contaminant per million parts of air (ppm) based on a 1-hour sampling period.

4.0 Training
The PI is responsible for ensuring individuals who handle Isoflurane have read the Safety Data Sheet (SDS) and receive training on the hazards of isoflurane regarding:
- Proper handling, use, storage, and disposal of Isoflurane
- The anesthesia procedures, including the use of the anesthetic machine and the waste anesthetic gas scavenging system
- Proper use of chemical fume hoods or other applicable local exhaust ventilation
- Spill clean-up and emergency response procedures

Training may be achieved by reviewing this guideline.

5.0 Personal Protective Equipment
Use personal protective equipment (PPE) routinely required for activities related to animal work, such as gloves and lab coats. When handling liquid solutions chemical splash goggles should be worn to protect against potential splashes. Additional PPE may be required based on the type of agents and procedures being conducted.

6.0 Equipment Checks and Maintenance
Each piece of equipment involved in the delivery of inhalant anesthetics, and removal of waste gases, must be evaluated prior to each use to assure its proper function and integrity. Tubing, hoses, and rubber items are common areas of concern and should be checked for leaks thoroughly before each use.

Anesthesia machines must be maintained in good working condition to assure optimal agent delivery in a safe manner. The primary standard for safety certification is the manufacturer recommendation. If such recommendation does not exist, use this guidance.

If following the manufacturer recommendation, a copy of the manufacturer service manual or instructions or certification requirements should be available within the laboratory to assist in adequate Institutional Animal Care and Use Committee (IACUC) oversight of anesthetic equipment.

If no manufacturer recommendation is available, then the equipment certification must be validated annually or any time the vaporizer has not been in service for more than a year.

Vaporizers must have documentation of safety certification & service. Information that must be maintained includes:
- Date of last service
- Date of the safety certification test
- Initials for the person who performed the test
- Test results
- Vaporizers should have a certificate of the safety certification date affixed to it.

Documentation of service must be affixed to each anesthesia machine or vaporizer that is in service.
7.0 Scavenging Equipment

Potential exposure routes include inhalation and skin contact. Waste anesthetic gas (WAG) must be scavenged through active or passive means. Active scavenging involves an active airflow system that draws WAG away from the researcher into an in-house WAG exhaust line, chemical fume hood, canopy hood or snorkel. Passive scavenging methods involve passing the WAG through an activated charcoal canister, after which it is discharged into the room. Scavenging equipment must be maintained in good working order to ensure a safe working environment. Approved options for scavenging equipment are:

- **Dedicated exhaust system**: A dedicated exhaust system such as an active vacuum waste gas line or an “elephant trunk” exhaust system is the preferred method to remove waste gases from the work environment.

- **Non-circulation ventilation systems**: non-circulation ventilation systems discharge waste gases through an exhaust vent or grill (e.g., hard-ducted biosafety cabinet or downdraft table).

- **Chemical fume hood**: The anesthetic can be delivered to the animal while it is inside the fume hood, or an exhaust gas line from the anesthesia machine can be vented inside the hood.

- **Adsorption devices**: Charcoal canisters such as F/Air or Enviro-Pure can be used to adsorb halogenated waste gases. These canisters must be properly positioned so that the vent holes on the bottom of the canister are not obstructed. Usage must be documented and accompanied by the method used to determine canister life as supplied by the manufacturer. See Table 1 below for examples of manufacturer’s recommendations. Always follow the latest recommendations from your canister’s manufacturer.

<table>
<thead>
<tr>
<th>Canister</th>
<th>Maximum Hours of Use</th>
<th>Maximum Weight Gain</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breath Fresh</td>
<td>12-15 hours</td>
<td>50 grams</td>
</tr>
<tr>
<td>F/Air</td>
<td>12-15 hours</td>
<td>50 grams</td>
</tr>
<tr>
<td>Enviro-Pure</td>
<td>N/A</td>
<td>100-120 grams</td>
</tr>
<tr>
<td>VaporGuard</td>
<td>N/A</td>
<td>50 grams</td>
</tr>
<tr>
<td>VetOne Clean Air Filter</td>
<td>12-15- hours</td>
<td>50 grams</td>
</tr>
</tbody>
</table>

8.0 Charcoal Canister Use

- Charcoal filters have a finite effective life span, which can be monitored by time in use, or weight. The weight of each new canister must be recorded before its first use.

- Before each use, the weight must be checked and recorded.

- After each use, record the amount of time in use. Do not exceed the maximum use hours of each canister as recommended by the manufacturer.

- To function properly, ensure that the charcoal canister is oriented in the proper position as recommended by the manufacturer.

- To function properly with adequate air flow, ensure that the holes (if applicable) on the bottom of the charcoal canister are not blocked.
9.0 Exposure Procedures

**Eyes:** If anesthetic gases/liquids come into contact with eyes, immediately flush them with copious amounts of water for at least 15 minutes using the emergency eyewash. If irritation persist or signs of toxicity occur, seek medical attention.

**Skin:** In the event of skin exposure, remove contaminated clothing and immediately wash the affected area with soap and water. If irritation persist or signs of toxicity occur, seek medical attention.

**Inhalation:** Move individuals to area with fresh air. Call 911 if any of the acute exposure symptoms are noted or if medical attention is necessary.

10.0 Spills

For small spills (a few milliliters), isoflurane is a highly volatile liquid and may dissipate quickly before cleaning up can be initiated. To minimize exposure, avoid attempting to clean up residual liquid and allow the isoflurane to evaporate. If attempting to clean up the spill, follow these steps:

- Wear appropriate personal protective equipment (PPE).
- Review cleaning/spill procedures found in the Safety Data Sheet (SDS)
- Absorb any residual solution with absorbent material (spill pads, paper towels, etc.) and place in fume hood for safe evaporation
- Dispose of absorbent material following waste disposal procedures below
- Clean spill area with soap and water

For large spills (a stock bottle):

- Do not attempt to clean up the spill
- Advise and warn co-workers
- Evacuate the area immediately
- Restrict access to the area
- Notify EHSO Spill Team at (404) 727-2888, providing the following information:
  - Location of spill
  - Name of hazardous material
  - Quantity involved
  - Any related health risks or exposure

11.0 Waste Disposal

Unused (expired) solutions of all anesthetic liquids need to be disposed of as a hazardous chemical waste through EHSO’s Environmental Group. To schedule a pick-up, click on this link [https://emory-integration.zerionsoftware.com/chemical_pickup](https://emory-integration.zerionsoftware.com/chemical_pickup).

Spent charcoal canisters should be sealed in a plastic bag, properly labeled, and submitted to EHSO as hazardous waste through the link above. **Do not** discard in the conventional trash.

12.0 Contact Information

For questions regarding this guidance, contact EHSO at indhyg@emory.edu. For questions regarding animal related research or anesthetic use approvals contact IACUC at iacuc@emory.edu.
13.0 References

OSHA Anesthetic Gases: Guidelines for Workplace Exposures
NIOSH Anesthetic Gases