

NIAAA Office of Laboratory Animal Science

ANESTHESIA MACHINE HANDBOOK

This information is intended for NIAAA DICBR animal users using the isoflurane anesthesia machines present in the FLAC (see page 3). For additional information on rodent anesthesia, machine set-up, testing, and troubleshooting see the manufacture's web site (e.g. <http://www.vetequip.com>).

Before anesthetizing animals with isoflurane:

- a. Ensure the level of isoflurane in the vaporizer is between the minimum and maximum level marks on the vaporizer. When adding isoflurane BE SURE the Oxygen Tank Valve is closed, the vaporizer is turned off, and there is no pressure in the system. Failure to do so may result in an eruption of isoflurane spray when opening the Isoflurane Fill Valve.
- b. Begin the supply of Oxygen to the anesthesia machine by turning the Oxygen Tank Valve counter clockwise $\frac{1}{4}$ of a turn. Check the Oxygen Tank Pressure Gauge and ensure there is enough oxygen remaining in the tank for the duration of the procedure(s). The gauge should be between 800 and 2300 psi. **Never BEGIN a procedure with the Oxygen Tank Pressure gauge in the red.**

Note: The tank pressure regulator is factory preset to deliver 50 psi which is the design pressure for the FLAC's precision vaporizers. No adjustment is necessary or desired.

Anesthetizing the animal: Place the animal in the induction chamber and latch the lid. Set the Oxygen Flow Meter to deliver one liter per minute (LPM) to the induction chamber. Set the vaporizer to deliver an isoflurane concentration of 2-4%. The animal should become unsteady within 90 seconds and anesthetized within 3-4 minutes.

WARNING: The induction chamber is airtight. If the oxygen flow meter on the machine is turned off OR the supply valve to the induction chamber is "OFF", there will be NO AIRFLOW into the chamber. NEVER leave animals in an induction chamber without an oxygen supply.

Once the animal's breathing is slow, moderately deep, and steady, the chamber should be "flushed" with oxygen by depressing (or "flipping") the oxygen flush valve for 20-30 seconds to flush the isoflurane out of the chamber before opening it to remove the animal. The animal should immediately be placed in the nose cone or stereotaxic frame and the oxygen flow rate decreased to provide $\frac{1}{2}$ LPM and the vaporizer setting decreased to 2% isoflurane. The level of anesthesia should be monitored and adjusted as necessary to prevent the animal from becoming too deep (very slow, very deep breaths with periods of apnea), or too light (rapid shallow breathing with whisker, limb or eye lid movements) during the procedure. Adjusting the % isoflurane in the anesthetic mixture adjusts the DEPTH of anesthesia. Adjusting the oxygen flow rate influences the RATE at which the subject's anesthesia level changes in response to changes in isoflurane concentration. Decreasing the Isoflurane concentration from 2 $\frac{1}{2}$ to 2 will "lighten" the animal's level of anesthesia. Simultaneously increasing the oxygen flow rate from 0.5 LPM to 2 LPM temporarily will achieve the lighter level of anesthesia more quickly. Simply increasing or decreasing the oxygen flow rate should not influence the level of anesthesia. ***IF*** it does, it is a sign of a leak in the system, excessive dead space in front of the animal's nose, or other issue that should be identified and resolved. 0.5 LPM for each animal in a nose cone or stereotactic circuit and 1.0 LPM for each animal in an induction chamber should be plenty of oxygen flow to meet the animal's oxygen and anesthesia needs. Any perceived failure of any of the machine components to perform as expected should be reported to the Facility Manager.

Recovery: In most cases anesthesia may be discontinued by shutting off the oxygen flow at the flow meter and placing the animal in a recovery cage. If the animal has had a long procedure, is hypothermic, or deeply anesthetized, the animal should be left on the anesthesia machine, the vaporizer turned to 0% or OFF, and the breathing circuit flushed with oxygen to clear residual isoflurane, and the oxygen flow meter left on at ½ LPM. Once the animal begins to move its whiskers or legs, the oxygen flow may be turned OFF and the animal removed from the nose cone and placed in a recovery cage. “Normal” recovery time (turning off the vaporizer, placing the animal in a recovery cage, the animal regains its righting reflex and begins walking) should be < 3 minutes. If recovery takes greater than 5 minutes, either an injectable anesthetic such as ketamine/xylazine has also been administered or something is wrong. If no injectable anesthetic has been administered, hypothermia is likely the issue. Refer to the Special Care Instructions for [Hypothermia](#).

Conclusion of the day’s procedures: When turning off the Oxygen Flow Meter, it is only necessary to turn the flow control knob clockwise until the ball float reaches the bottom of the tube. Excessive tightening will damage the valve seat, resulting in a leak requiring replacement of the valve. After turning off the flow meter, the Oxygen Tank Valve should be turned off and residual pressure relieved from the system. The induction chamber should be cleaned and sanitized with the quaternary ammonium detergent disinfectant provided. Any worn or torn face mask membranes should be replaced.

“Thumb” rules for mice

Acceptable oxygen tank operating pressures:
800-2300 PSI

Oxygen flow rate:
0.5 LPM for face mask or stereotaxic frame
1-2 LPM for induction chamber

Signs of a “Surgical plane” (Stage 3, Level 2 depth) of general anesthesia:
lack of blink or pedal reflex
12-25 medium deep steady breaths / minute
Heart rate = 250-450 / minute

Body temperature:
Normal awake = $36.5 \pm 1^\circ \text{C}$
Anesthetized = $34\text{-}36.5^\circ \text{C}$
Severe hypothermia = $< 32^\circ \text{C}$
Life threatening hypothermia = $< 28^\circ \text{C}$

Oxygen Tank Pressure Gauge



Oxygen Tank Valve

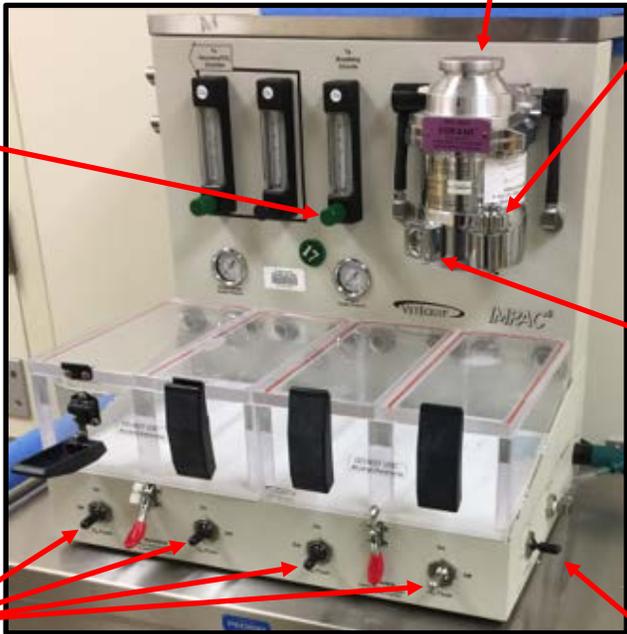
Oxygen Flow Meter



Isoflurane Vaporizer concentration adjustment



Isoflurane Fill Valve



Isoflurane level



Induction chamber Oxygen flush valve



Face mask Oxygen flush valve

