

COMPARATIVE MEDICINE
LABORATORY ANIMAL FACILITIES

STANDARD OPERATING PROCEDURE
for
CO₂ CHAMBER OPERATION

- 1.0 Purpose:
This procedure provides operating instructions for various CO₂ chambers throughout the CMLAF.
- 2.0 Scope:
This procedure applies to all CMLAF and principal investigator staff.
- 3.0 Procedure:
 - 3.1 Open the CO₂ tank valve. Check pressure gauge. Switch tanks if more CO₂ is needed.
 - 3.2 Open door to the CO₂ chamber.
 - A. Obtain keys to unlock from area supervisor or main office at BEB.
 - 3.3 Place animals in the chamber. Close door.
 - 3.4 Slowly turn the regulator valve clockwise until gas is heard entering the chamber.
Note: When the regulator valve is open, the CO₂ stream flowing through it cools very rapidly. Watch for the formation of "dry ice" between the regulator valve and the euthanasia chamber. If ice begins to form, reduce flow into the regulator valve, or, if necessary, turn off the regulator valve until the ice melts.
 - 3.5 Add CO₂ to the chamber SLOWLY by turning the regulator valve. This allows air and CO₂ to mix, and results in the gradual unconsciousness of the animal. Do not allow sudden exposure to 100% CO₂ because it will cause asphyxiation and distress.
 - 3.6 Allow the animals to remain in the CO₂ chamber for at least five minutes.
 - A. Observe carefully to ensure that the animals are dead before removing them from the chamber.
 - B. Neonatal animals appear to be resistant to hypoxia and are able to tolerate relatively high CO₂ concentrations. Decapitation (with scissors) must follow CO₂ narcosis for neonates less than seven (7) days old.
 - 3.7 Ensure animals are dead before disposal.
 - A. Often animals can resuscitate when re-exposed to room air.
 - B. Palpate chest for heart beats to make sure the heart has stopped.
 - C. Cut open chest (thoracotomy) with scissors or scalpel before disposing carcasses in freezer.
 - 3.8 When finished, be sure the regulator valve is turned off.
 - 3.9 A CO₂ back up tank will be available at all times.

- A. To avoid interruptions, check tank pressure gauge after each procedure.
 - 1. When pressure reaches 600 PSI or less, notify floor supervisor or call the CMLAF main office at 829-2919 to let them know that a new tank needs to be ordered.
- 3.10 The need to minimize animal distress, including fear, anxiety and apprehension must be considered at all times. Therefore, euthanasia CANNOT occur in animal holding rooms.
- 3.11 Animals awaiting euthanasia must be located as far away as possible from those actually undergoing euthanasia.
- 3.12 CO₂ chambers can be used by any principal investigator with protocol approval, or with proper authorization of the attending veterinarian.
- 3.13 Principal investigators should be instructed to make an appointment with a technician or the area supervisor on how to properly use the chamber.