RODENT EUTHANASIA INSTRUCTIONS

Building Supplied CO₂

For more detailed step-by-step instructions, please refer to the Guidelines for Rodent Euthanasia Procedures for Investigative Personnel (animalcare.umich.edu/rodent-euth)
CAGES MUST BE LABELED WITH:

- PI Name
- Protocol Number
- Shortcode
- Lab Contact Information

Until euthanasia is confirmed by secondary, physical means
REMEMBER, IF CONSOLIDATING MICE OR RATS FOR EUTHANASIA:

- **Do not consolidate rodents** except for mice or rats

- **Follow standard housing densities** when consolidating rodents (except mice) for euthanasia

- **No more than** 10 mice in a standard mouse (#1) cage or 25 mice in a large mouse / small rat (#3) cage

- Provide ample space for animals to move and assume normal postures if using your own euthanizing chamber

- Start the procedure **no more than 10 minutes** after the animals have been consolidated
The flow meter (1) should be at zero.

The supply valve (2) and flow meter (3) should be turned off (all the way clockwise).
STEP 02

Insert the CO$_2$ hose through the water bottle grommet on the cage lid

OR

through the lixithole in the back of the cage
Turn supply valve (1) to the left or counterclockwise to turn on the CO₂
Adjust CO$_2$ flow into the cage by turning regulator knob (1) slowly

Until the gauge (floating black ball) (2) reaches the desired color line
Allow gas to flow until animals are no longer moving or breathing

You **MUST** remain with your animals until death is confirmed by a secondary means
Turn off the CO\textsubscript{2} supply valve (1)

Note that gauge (2) is falling back to zero now
Gauge should now be set at zero with no more CO₂ flowing.

After gauges reach zero ensure flow meter valve (1) is shut off by turning it clockwise until snug.

Do NOT overtighten.
Check your protocol for approved secondary euthanasia method

**ALWAYS** perform a secondary, physical method to ensure euthanasia is effective
After you have confirmed death by a secondary, physical means:

Put carcass in a glove or opaque bag.

Label with PI name, date, and name of individual conducting euthanasia.
Place in appropriate bin within carcass cooler
We want to acknowledge that the ethical cost of using animals in research may cause some individuals emotional distress, especially if they perform euthanasia as part of their occupational/research duties.

If you think you may be struggling with what is commonly referred to as “compassion fatigue,” there are resources that can help.

Visit animalcare.umich.edu/cap or email compassionawareness@umich.edu to learn more about our Compassion Awareness Project.