For more detailed step-by-step instructions, please refer to the Guidelines for Rats & Mice Euthanasia Procedures for Investigative Personnel (az.research.umich.edu/node/1016)
REMEMBER, IF CONSOLIDATING ANIMALS FOR EUTHANASIA:

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

Follow standard housing densities when consolidating other rodents (e.g. rats) for euthanasia

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
Tank valve (3) should be turned off
Pressure gauge (1) should read zero
Flow meter (2) should read zero
Place euthanasia lid over top of cage

Remove MI-top, feeder, and water

If no euthanasia lid present, CO$_2$ hose can be inserted directly through water bottle grommet or water valve opening
Turn tank valve (2) to the left (counterclockwise) until the needle rises on the pressure gauge (1).
Adjust CO₂ flow into the cage by turning flow meter knob (1) 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 CO₂ by turning the tank valve (3) clockwise until it is snug.

Gauges 1 and 2 should fall back to zero.
Both gauges should now be at zero with no CO₂ flowing
Turn the CO$_2$ flow meter valve until snug in the off position

Do NOT over tighten
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 CompassionFatigue@umich.edu to learn more about our Compassion Awareness Project.