

Gas cylinders – Standard operating procedure

Updated 2018-05-29 (Roger Shih)

PURPOSE

The lab uses carbon dioxide gas for cell-culture incubators, and nitrogen gas to spray dust off of glass slides, e.g. prior to coating them. This document provides instructions for safe usage of compressed gas cylinders and air guns.

PERSONAL PROTECTIVE EQUIPMENT (PPE)

- Safety glasses/goggles

PROCEDURE

Carbon dioxide cylinders remain connected to the incubator at ~10 psi, and are not adjusted until they run low and need swapping. Nitrogen cylinders are opened and closed more frequently, to conserve them while not in use.

There are three knobs on a gas cylinder and regulator (Fig 1):

- **Knob 1** opens and closes the gas cylinder. In typical usage, the other two knobs do not need to be adjusted. Simply open knob 1 slowly before use (a partial turn is sufficient), and close it when finished.
- **Knob 2** controls the regulator's output pressure. For simplicity, this is only adjusted when swapping tanks, and is otherwise left alone. Carbon dioxide tanks are set at ~10 psi, as mentioned. Nitrogen is set between 30-60 psi. Contrary to most knobs, screwing in (clockwise) raises the pressure, and screwing out (counterclockwise) lowers the pressure. Screwing out too much will detach the knob from the regulator, but it can simply be screwed back on.
- **Knob 3** opens and closes the regulator output. This is typically left open.

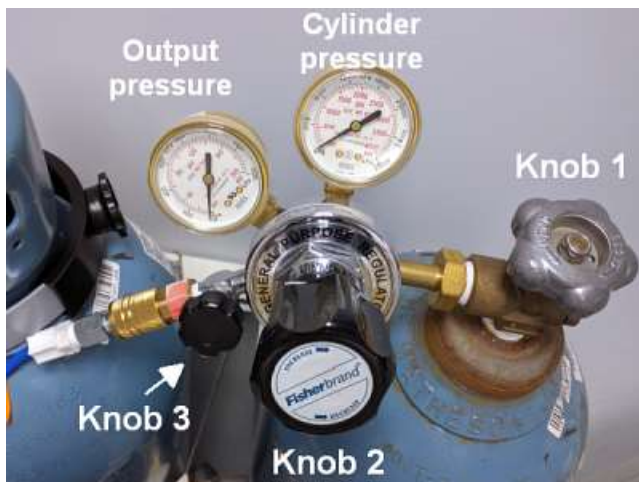


Fig 1: Knobs and gauges on cylinder and regulator. **Fig 2:** Air gun.

The air gun (Fig 2) is simply controlled by squeezing the blue lever to spray gas. When spraying a target such as a glass slide, beware not to blow the object out of your grasp. There is a filter in the barrel, which can be periodically replaced by unscrewing the barrel.

If the cylinder runs low on gas, retire it before it is completely empty. (Leaving a little pressure inside helps prevent water vapor entry and internal rusting.) Contact the lab member in charge of maintaining the gas tanks; they will transfer the pressure regulator to a new tank and order a swap for the empty tank.

HAZARDS

High pressure: Do not adjust knob 2's regulator pressure higher than necessary (e.g. 60 psi for nitrogen). Do not spray gas into face or on exposed skin. Do not spray near lightweight objects, and grip target with appropriate firmness while spraying. Do not unstrap cylinder from wall fixture.

Asphyxiation: Gas leakage and buildup can lower the local oxygen concentration, causing sudden loss of consciousness, followed by death if not promptly moved to fresh air. (As always, do not work alone in the lab.) Whereas carbon dioxide causes telltale suffocating sensations as it builds up, nitrogen asphyxiation triggers no bodily warnings even as oxygen levels drop. If you hear a cylinder leaking while the air gun is not spraying, close knob 1 and let labmates know not to use the cylinder until it can be inspected.

RESOURCES

Chemistry department compressed-gas safety
https://www.chem.utoronto.ca/safety/compressed_gases.php

EH&S Chemical and Lab safety (see Compressed Gas subpage)
<https://ehs.utoronto.ca/our-services/chemical-and-lab-safety/>

APPENDIX: SWAPPING REGULATORS

When a cylinder runs low, the regulator needs to be transferred to a new tank. For safety, **do not attempt this without experience**. Instead, contact the designated lab member in charge of gas cylinders. Steps are documented below:

1. Close knobs 1, 2, and 3, in that order.
2. Use a wrench to unscrew the regulator from the empty cylinder.
3. Cap the empty cylinder.
4. Label the empty cylinder as "empty".

5. Uncap the new cylinder.
6. Screw the regulator into the cylinder with a wrench. Do not overtighten.
7. Slowly open knob 1 and listen for leakage (hissing) from the fitting.
8. If there is leakage, close knob 1, unscrew the regulator from the cylinder, and replace the white Teflon tape on the regulator screw threads. One layer should suffice. (In theory, fittings are designed to not need Teflon tape for an airtight seal, but this is not always the case in practice.)
9. Repeat steps 6-8 until fitting is airtight.
10. Adjust knob 2 to appropriate pressure: 10 psi for carbon dioxide, or 30-60 psi for nitrogen.
11. Open knob 3.
12. For carbon dioxide, leave all knobs open. For nitrogen, briefly test air gun, then close knob 1.