

## Using the Carbon Dioxide Incubator in the Digital Microscopy Center

### Set up and usage.

Turn on the incubator:

1. Turn on power by turning the key to the first position, "I".
  - a. this position turns on the incubator without an alarm
  - b. the last position activates a low CO<sub>2</sub> alert.

If carbon dioxide (CO<sub>2</sub>) is required:

1. Turn on tank interchange system using the black switch on the left front;
2. Open the CO<sub>2</sub> supply:
  - c. check that each regulator's knob has been turned fully counter-clockwise
  - d. open both CO<sub>2</sub> tanks by turning their valves fully counter-clockwise
  - e. set both regulators to 15 psi by slowly turning their knobs clockwise until the low pressure gauge needle (left hand gauge) points to 15-18 psi

Turn off.

1. Turn off power to the incubator by turning the key to "0";
2. Turn off power to the interchange system by the black switch on its front panel
3. Turn the knob on both regulators fully counterclockwise;
4. Close the CO<sub>2</sub> cylinders by turning their valves fully clockwise.

### Do you need CO<sub>2</sub>?

Zebrafish do not need CO<sub>2</sub>.

Cultured mammalian tissues only need CO<sub>2</sub> if they are in carbonate-buffered culture media. Culture media containing non-carbonate buffers such as HEPES or PIPES do not require CO<sub>2</sub>.

### Correct use of regulators – "Why do I have to turn the knobs?"

Gas regulators have 2 gauges, the high pressure gauge and a low pressure gauge. The high pressure side of the regulator can withstand the tank's full pressure, up to 2500 pounds per square inch (psi) or nearly 200 times atmospheric pressure. The low pressure side of the regulator can easily be damaged when the tank's valve is quickly opened.

The right hand, high pressure gauge on the regulator indicates how much pressure remains in the gas cylinder. The left hand gauge indicates how much pressure is leaving the regulator.

The regulator knob consists of a brass bar forming a "T" extending out from the body of the regulator.

Turning the regulator knob fully counterclockwise prevents pressure from the gas cylinder from reaching the regulator mechanism. The knob is turned slowly clockwise after the tank is opened until the desired amount of output pressure is indicated on the low pressure gauge.

### Tank Interchange System.

This ensures that the CO<sub>2</sub> incubator does not run out of CO<sub>2</sub> gas. It feeds gas from one cylinder to the incubator and when the feed cylinder is empty, the interchange device switches to the standby cylinder. At this point, note which cylinder is empty and make arrangements to replace it with a full cylinder (usually, notify DMC staff).

A green light indicates which gas cylinder is in use. The other cylinder is on standby. A flashing red light indicates that a cylinder is empty and needs to be replaced.

### Setting the incubator temperature.

Three buttons are present below the incubator's display screen, Increase, Mode, Decrease.

1. Turn the incubator key to the first position;
2. The Revco logo will display on the incubator screen;
3. While the logo is displayed, press and hold the Increase button until the logo disappears, then release the button;
4. Press the Increase or Decrease buttons to change the blinking parameter;
5. Press the Mode button to accept the displayed value and move to the next parameter;
6. If you do not adjust the flashing parameter within 60 sec, the software will accept the value and move to the next parameter.

Adjustable parameters (default):

Temperature (37° C)

Warm alarm (40° C)

Cold alarm (34° C)

CO<sub>2</sub> (5% default)

High CO<sub>2</sub> alarm (6%)

Low CO<sub>2</sub> alarm (4%)

The manufacturer's manuals for the incubator, interchange system and regulators are located in the CD175 filing cabinet