

## **INSTRUCTIONS FOR HOT BOX SYSTEM™**

The HOT BOX™ system consists of four major components. First, the HOT CHIMNEY™ which removes the radioactive volatile, generated during storage, from the reagent vial. Second, the HOT FILTERS™ which tightly binds the radioactive volatile generated during experimentation. Third, the HOT BOX™ (Modular Incubator Chamber™) serves as an incubator and containment vesicle. Finally, the flow meter this regulates the flow of gases to ensure operational safety and maximal retention time for removal of radioactive volatile. The dual flow meter is also used to mix two different gases.

**FOR MAXIMUM SAFETY, RETENTION TIME AND EFFICIENCY DO NOT EXCEED A FLOW RATE GREATER THAN 7 LPM AND A PRESSURE OF 2 PSI.**

**NOTE: ALL OF THE FOLLOWING OPERATIONS SHOULD BE CARRIED OUT IN A WELL VENTILATED HOOD.**

### **AIR FILTERS**

One air filter is provided with each unit and is used to keep flow meter clean. It is attached between gas source and inlet (bottom barbed brass fitting) of flow meter.

### **HOT BOX™ AND ATTACHMENT OF HOT FILTERS™**

**To familiarize yourself with the proper use of the HOT BOX™, please read and follow INSTRUCTIONS FOR MODULAR INCUBATOR CHAMBER™.**

B1. Place experiment containing 35S-labeled reagent in a HOT BOX™ (round 12 in. clear polycarbonate chamber). The chamber can be **humidified** by placing a petri dish filled with 10 ml of sterile water at the bottom of the chamber. The water in this dish will absorb a small percentage of the 35S volatile.

B2. Place one static HOT FILTER™ (6 in. diameter disk) onto the filter post. Filter should rest on the top ledge of post. (If using 150mm petri dishes remove post and place static filter on top of petri dish or in a tray above experiment.)

B3. Place chamber lid on base.

B4. Put stainless steel clamp on the chamber at position where base and lid join. Left hand should be held firmly against clamp and chamber. Slowly close clamp handle with right hand making sure clamp is centered.

B5. **Check to see that white tubing clamps of gas inlet and outlet ports are in OPEN POSITION. Failure to do so will cause excessive back pressure to build up in the system.**

B6. Insert first 1/4 of the **small end** of EXHAUST HOT FILTER™ (Black tube) to tygon tubing of either port. Do not insert it too far into the tygon tubing, otherwise it will be difficult to remove later. The best way is to line up the end of the tygon tubing with the first barb mark on the EXHAUST HOT FILTER™.

### **FLOWMETER AND ATTACHMENT OF THE FLOWMETER TO THE HOT BOX™**

C1. Open regulator of gas tank to allow minimum gas flow.

C2. Attach the flow meter inlet tube (bottom tube) to gas tank and adjust both the flow meter and regulator on the gas tank to allow 5-7 liters per minute (LPM) of flow rate.

C3. Connect gas outlet tube (top tube) of the flow meter to the Hot BOX™. Flush the system for 8-10 minutes at a flow rate of 7 LPM. Pressure release valve in-line with the

flow meter will open if functional and safe flow rate is exceeded. The pressure release valve will automatically close when pressure is safe.

C4. Disconnect the HOT BOX™ from the flow meter.

C5. Seal HOT BOX™ by closing plastic clamps (WHITE) of gas inlet and outlet ports on the BOX.

C6. Remove the EXHAUST HOT FILTER™ (BLACK TUBE) from the HOT BOX™ and properly store for use at the end of the experiment.

C7. Put HOT BOX™ at appropriate temperature either in a conventional continuous flow CO<sub>2</sub> incubator, an oven, or a warm room.

C8. At the end of incubation but, before opening the HOT BOX™,

reattach the EXHAUST HOT FILTER™ to HOT BOX™ (see step B6).

C9. Open the gas inlet and outlet ports on HOT BOX by opening the white clamps.

C10. Repeat steps C1-C4 to remove any radioactive volatile not absorbed by the STATIC HOT FILTER™ inside the HOT BOX™ during labeling experiment.

C11. Remove both the EXHAUST and STATIC HOT FILTERS™ and dispose of them in radioactive waste.

C12. The water and petri dish used to humidify the HOT BOX™ will be radioactive and should be put in radioactive waste.

C13. Insides of HOT BOX™ and the attached tygon tubing should be monitored for counts. If counts become excessive, decontaminate the HOT BOX™.

C14. Seal and store HOT BOX™ until next experiment.

#### **Flow meter (Single or Dual)**

To start system, open the valve slowly to avoid possible damage. Rate of flow is read at the point of maximum horizontal width for spherical floats or at the top of the largest diameter for non-spherical floats. Control valves are turned clockwise to reduce flow, counter clockwise to increase flow. A nylon insert is provided in the threaded section of the valve stem to give a firm touch to the valve and to prevent change of setting due to vibration.

#### **INSTRUCTIONS FOR CONVERSION OF THE MODULAR INCUBATOR CHAMBER™ INTO A HOT BOX™**

To convert your present Modular Incubator Chamber™ into a HOT BOX SYSTEM™ follow the steps below.

#### **Filter Post (Threaded White Rod)**

Place filter post into center hole of bottom tray. Put white nut on and tighten by hand. One to three trays can be used. Always put the tray with post on the bottom of the chamber.