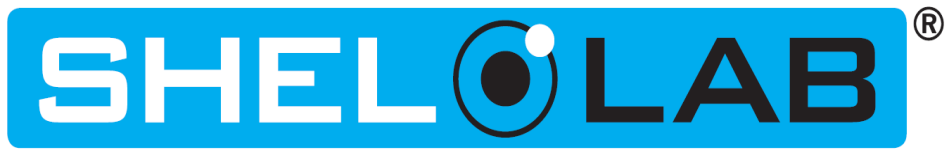


BACTROX 110 – 120 Volts



Installation and Operation Manual

BACTROX

BACTROX HYPOXIA CHAMBER 110 - 120 Volts

Installation and Operation Manual

Part Number (Manual): 4861714

Revision: May 21, 2015

These units are TÜV CUE listed as climatic chambers for professional, industrial, or educational use where the preparation or testing of materials is done at an ambient air pressure range of 22.14 – 31.3 inHg (75 – 106 kPa) and no flammable, volatile, or combustible materials are being heated.

These units have been tested to the following requirements:

CAN/CSA C22.2 No. 61010-1:2012

CAN/CSA C22.2 No. 61010-2-010:2004 Reaffirmed: 2014-07

UL 61010-1:2012-05

UL 61010A-2-010:2002-03

EN 61010-1:2010

EN 61010-2-010:2014

Supplemented by: UL 61010-2-010:2015

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INTRODUCTION

Thank you for purchasing a Shel Lab BACTROX Hypoxia Chamber Workstation. We know that in today's competitive marketplace, customers have many choices when it comes to constant temperature equipment. We appreciate you choosing ours. Our continued reputation as a leading laboratory product manufacturer rests with your satisfaction. Sheldon Manufacturing, Inc. stands behind our products, and we will be there if you need us.

BACTROX workstations are intended for professional, industrial, and educational applications as hypoxia workstations suitable for the cultivation of bacteria and cell cultures. They are not intended for use at hazardous or household locations. Only use this equipment within its intended spectrum of applications; any alterations or modifications void the warranty.

GENERAL SAFETY CONSIDERATIONS

Note: Failure to follow the guidelines and instructions in this manual may create a protection impairment by disabling or interfering with the unit's safety features. This can result in injury or death.

Before using the BACTROX read this entire manual carefully to understand how to install, operate, and maintain the workstation in a safe manner. Keep this manual available for use by all workstation operators. Ensure that all operators are given appropriate training prior to using the BACTROX.

The BACTROX and its recommended accessories are designed and tested to meet strict safety requirements. The workstation is built to connect to a wall power source using the specific power cord type shipped with the unit.

For safe operation of your BACTROX, always follow basic safety precautions including:

- Follow all local or regional ordinances in your area regarding the use of this unit. If you have any questions about local regulations, please contact the appropriate agency.
- Use only approved accessories. Do not modify system components. Any alterations or modifications to your BACTROX can be dangerous and void your warranty.
- Always plug the BACTROX power cord into an earth grounded electrical outlet that conforms to national and local electrical codes. If the workstation is not grounded properly, parts such as knobs and controls can conduct electricity and cause serious injury.
- Avoid damaging the power cord. Do not bend it excessively, step on it, or place heavy objects on it. A damaged cord can be a shock or fire hazard. Never use a power cord if it is damaged.
- Position the workstation so the end-user can quickly unplug the unit in the event of an emergency.
- Do not attempt to move the workstation while it is in operation.

INTRODUCTION (CONTINUED)

ENGINEERING IMPROVEMENTS

Sheldon Manufacturing continually improves all of its products. As a result, engineering changes and improvements are made from time to time. Therefore, some changes, modifications, and improvements may not be covered in this manual. If your unit's operating characteristics or appearance differs from those described in this manual, please contact your Shel Lab dealer or distributor for assistance.

CONTACTING ASSISTANCE

If you are unable to resolve a technical issue with the BACTROX, please contact Sheldon Technical Support. Phone hours for Sheldon Technical Support are 6am – 4:30pm Pacific Coast Time (west coast of the United States, UTC -8).

Please have the following information ready when calling or emailing Technical Support: the **model number** and the **serial number**. These will be found on the unit's data plate. See the [Recording Data Plate Information entry](#) on page 10, for the location of the data plate.

EMAIL: tech@shellab.com PHONE: 1-800-322-4897 extension 4 or (503) 640-3000 FAX: (503) 640-1366

Sheldon Manufacturing INC.
P.O. Box 627
Cornelius, OR 97113

RECEIVING YOUR BACTROX

Before leaving our factory, all BACTROXes are packaged in high-quality shipping materials to provide protection from transportation-related damage. When the unit departs the factory, safe delivery becomes the responsibility of the carrier. Damage sustained during transit is not covered by the BACTROX warranty.

This makes it important that you inspect your BACTROX for concealed loss or damage to its interior and exterior when receiving it. If you find any damage to the workstation, follow the carrier's procedure for claiming damage or loss.

Carefully check all packaging before discarding. Save the shipping carton until you are certain that the unit and its accessories function properly.

INSPECTING THE SHIPMENT

Carefully inspect the shipping carton for damage. Report any damage to the carrier service that delivered the BACTROX. If the carton is not damaged, open the carton and remove the contents. The unit should come with an Installation and Operation Manual, warranty card, and a Certificate of Compliance. Verify that the correct number of accessories are included with the unit:

Arm Port Doors
Left and Right



Petrie Dish Rack
7 (2 x 11 Plates)



5110729

Gas Regulator, CO₂



9740558

Gas Regulator,
Nitrogen



9740546

Leveling Feet (4)*



2700506

Power Cord 5-15
NEMA



1800510

Rubber Cuff Plugs



7200502

Shelf Spacers (3)



5680502

Sleeve Cuff
Assemblies
2 (Size 9 Large)



9990738L

Calibration Ambient
Air Sample Pump



9490571

Calibration Kit Cover



9490570

Cal / Data Logging
RS232 Cable



0860525

Calibration RS232 to
USB Adaptor Kit



0860524

Document Shelf



*BACTROXes shipped mounted on a stand do not include level feet.

RECEIVING YOUR BACTROX (CONTINUED)

Inspect the BACTROX for damage.

ORIENTATION

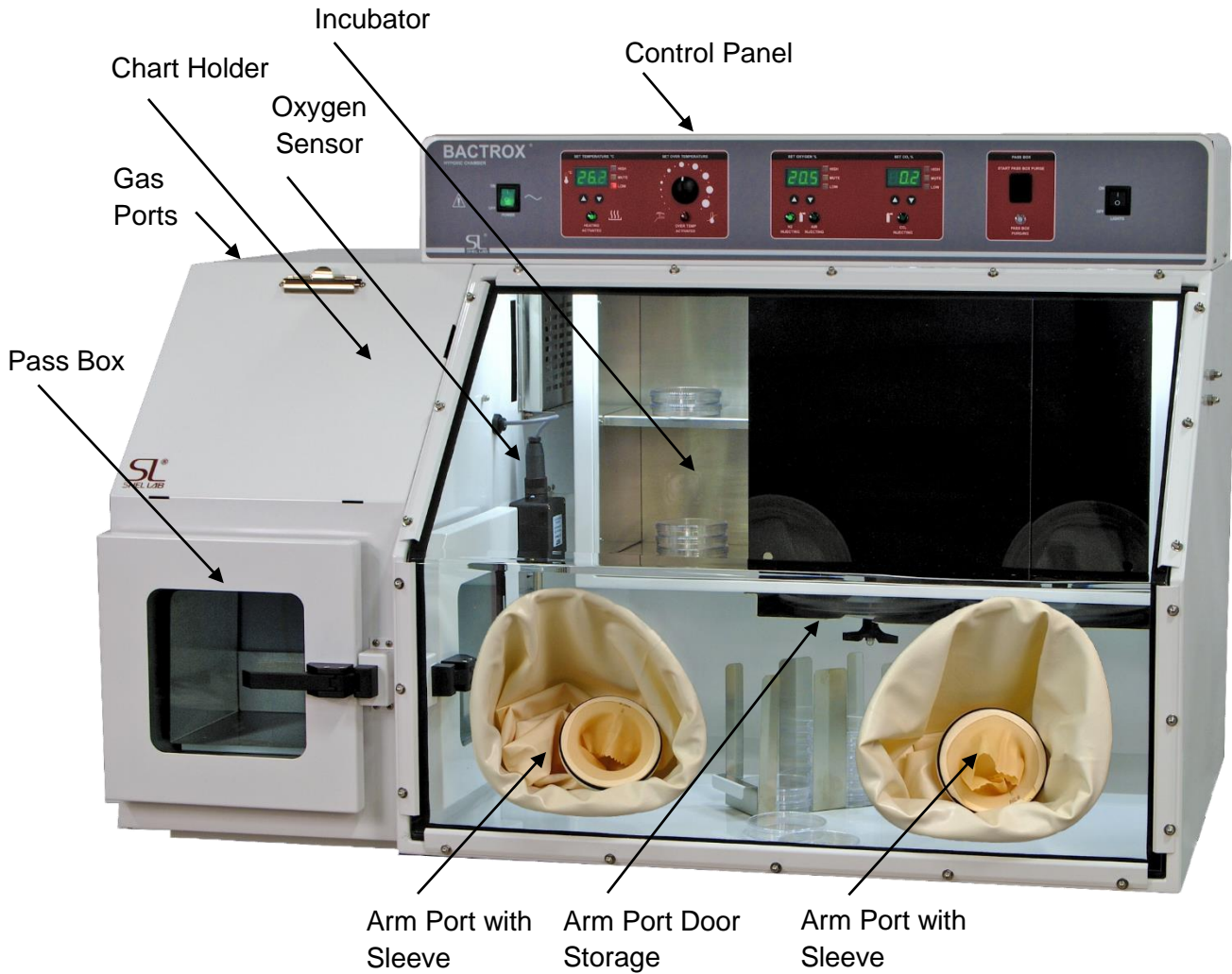


Figure 1: BACTROX Exterior

RECEIVING YOUR BACTROX (CONTINUED)

Additional Chamber Orientation



Figure 3: O₂ Sensor

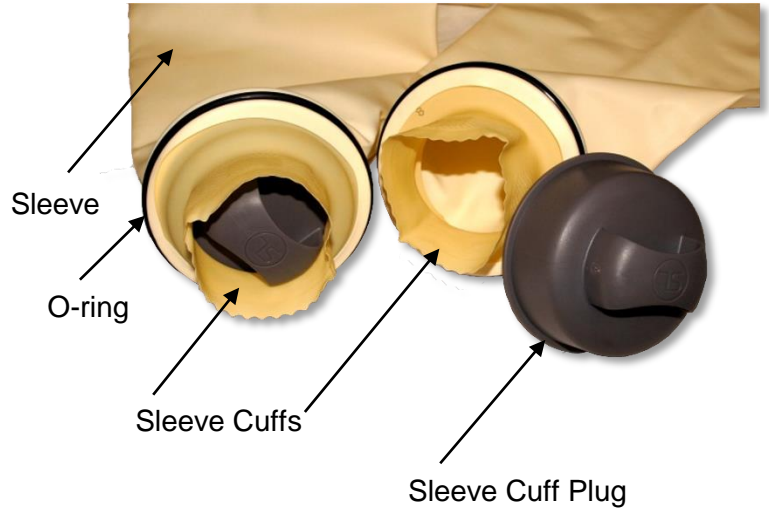


Figure 2: Sleeve Assemblies

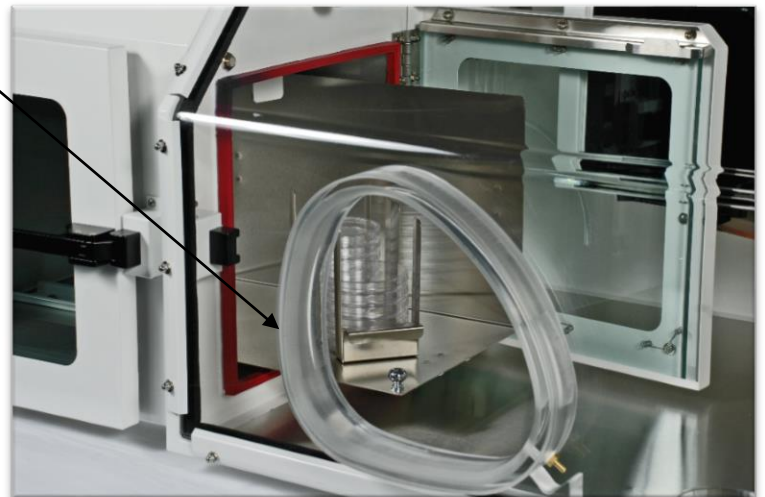
Oxygen Sensor

Power Outlet. Note:
Outlet type will vary
by voltage and nation.

Sensor Box

Pass Box Sliding
Shelf

Figure 4:
Pass Box



RECEIVING YOUR BACTROX (CONTINUED)

Vents – Back of the Unit



Figure 6: Chamber Vent

Oxygenated atmosphere is forced out of the workspace chamber through this vent. Do not plug or obstruct!



Figure 5: Pass Box Vent

During pass box purge cycles evacuated air is exhausted through this vent. The pass box vent should never be plugged while the unit is in operation. Remove the vent cover after installing the unit.

RECORDING DATA PLATE INFORMATION

Locate the data plate in the workspace chamber above the inner pass box door. The data plate contains the BACTROX model number and serial number. Enter this information below for future reference.

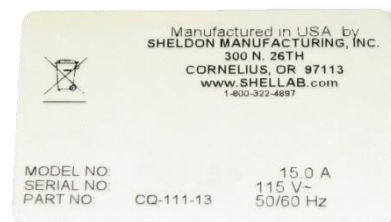


Figure 7: Data Plate

Date Plate Information

Model Number	
Serial Number	

RECEIVING YOUR BACTROX (CONTINUED)

CALIBRATION ITEMS

Calibration Gas – O₂ Display and Sensor

The BACTROX oxygen sensor requires periodic calibrations to compensate for drifts in accuracy. These drifts are caused by a naturally occurring material evolution over the operational lifetime of the sensor. The BACTROX O₂ sensor can be calibrated to a single O₂ value with a matching sample. However, a dual point calibration, calibrating at high value and then a low value, provides accuracy across a range of O₂ concentrations. The high value sample may be pulled from the ambient atmosphere, after calculating its oxygen concentration (please see page 84). Since the low value for a hypoxic range must be below the atmospheric concentration, a man-made supply with a known hypoxic O₂ concentration is required. Single point calibrations for hypoxic values also require a manufactured sample gas.

Such reference or calibration gases are purchased in a supply cylinder, which will require its own regulator and tubing to connect to the BACTROX. Check with your gas supplier for the appropriate regulator type. Only use oxygen – nitrogen mixes for O₂ sensor calibrations. Do not use an O₂ - CO₂ mix for an O₂ calibration, even if you will be running a carbon dioxide-enriched environment.

O₂ sensor calibrations typically only use a small volume of gas.

Calibration mixes are sometimes known among gas suppliers as **Certified Standard Mixtures** or **Portable Calibration Gases** with a **% Oxygen**.

The BACTROX is dual-point calibrated at the factory. The low value is supplied with a certified standard gas mix purchased from Airgas with 5% O₂ – 95% N₂ balance (Airgas part number X02NI95C3006050). Airgas also provides other certified O₂ – N₂ mixes suitable for accurately calibrating the BACTROX at a variety of hypoxic values.

These include:

1% O ₂ - 99% N ₂	8% O ₂ - 92% N ₂
3% O ₂ - 97% N ₂	10% O ₂ - 90% N ₂
5% O ₂ - 95% N ₂	21% O ₂ - 79% N ₂

CO₂ Calibration Reference Device

The carbon dioxide (CO₂) sensor operates at significantly cooler internal temperatures than the O₂ sensor, and does not require a calibration gas. A digital reference gas analyzer capable of detecting CO₂ concentrations at least of 0.1% will suffice, and must be purchased separately.

Incubator Temperature Reference Device

The incubator requires a temperature reference thermometer accurate to at least 0.1°C for performing temperature display calibrations. The reference sensor must be purchased separately. For best results, use a digital reference device with a wire sensor probe to take remote readings. Remote readings may offer an hour or more of time savings during each calibration. Do not use an alcohol or mercury thermometer.

INSTALLATION

INSTALLATION CHECK LIST

Perform the following procedures in the Installation section to prepare and install the BACTROX in a laboratory or workspace location.

- ✓ **Check Ambient Conditions**, page 13
 - Verify that the room temperature, humidity level, and space available fall within the BACTROX-required ranges.
- ✓ **Check Location**, page 12
 - Check that potential locations do not expose the BACTROX to significant sources of heat or cold.
- ✓ **Power Source**, page 13
 - Verify that potential locations have power sources that match the power requirements listed on the BACTROX data plate, inside the chamber.
- ✓ **UV Lighting**, page 14
 - Verify that potential locations do not expose the BACTROX to UV lighting or direct sunlight
- ✓ Read the **Lifting and Handling** and the **Leveling** entries, page 15
 - Read how to safely move and level the unit.
- ✓ **Gas Sources and Usage**, page 16
 - Read about gas source types, requirements, and N₂ usage rates.
- ✓ **Install the Workstation**, page 17
 - Install the BACTROX in a location that matches the criteria listed in the previous procedures.
- ✓ **Connect to Gas Sources**, page 17
 - Connect the BACTROX to its nitrogen gas source and any CO₂ source.
- ✓ **Cleaning and Install Accessories**, page 18
 - Remove shipping wrapping from and clean the BACTROX interior, shelving, and accessories.
 - Place the cleaned accessories inside the workspace chamber.
- ✓ **Shelving Installation**, page 19
 - Install the shelf spacers in the workspace incubator
- ✓ **Install Arm Port Doors and Install the Sleeve Assemblies**, page 19
 - Install the arm port doors in the arm ports on the front panel of the unit, then install the sleeve assemblies on the ports.
- ✓ Remove the **Pass Box Vent Cover**, page 21

INSTALLATION (CONTINUED)

CHECK AMBIENT CONDITIONS

This workstation is intended for use indoors at room temperatures between **15°C and 30°C (59°F and 86°F)**, at no greater than **80% Relative Humidity** (at 25°C / 77°F). Allow a minimum of 4 inches (10cm) between the workstation and walls or partitions, and 2 inches (5cm) of clearance above the top of the workstation for unobstructed airflow.

Operating the unit outside of these conditions may adversely affect the unit temperature range and stability.

For conditions outside of those listed above, please contact your distributor or Shel Lab representative to explore other unit options suited to your laboratory or production environment.

CHECK LOCATION

When selecting a location to install your BACTROX, consider environmental factors that can affect the workstation temperature stability and atmospheric integrity:

- Ovens, autoclaves, and any device that produces significant radiant heat
- Heating and cooling ducts, or other sources of fast moving air currents
- High-traffic areas
- Direct sunlight

Note: Direct exposure to air conditioning vents or other sources of cold air can result in condensation or fogging on workstation acrylic glass panels, depending on humidity and other ambient conditions. Prolonged exposure to cold air flows may adversely affect the temperature performance of the incubator.

POWER SOURCE

Always position the workstation so that the users have access to the power cord and can quickly unplug it in the event of an emergency.

When choosing a location for the BACTROX check that the voltage and ampere requirements on the workstation data plate match those of your wall source. The source must be an earth grounded outlet. **The supplied voltage must not vary more than 10% from the data plate rating. Damage to the workstation may result if supplied voltage varies more than 10%.**

These incubators are intended for 50/60 Hz, 110 - 120 volt applications at 3.5 Amps. A T type 4 amp fuse is included in the power cord plug inlet on the back of the unit.

Use a separate circuit to prevent loss of the unit due to overloading or circuit failure.

Each BACTROX workstation is provided with a 110 – 120VAC 8ft (2.5m) 5-15 NEMA power cord.

Note: Electrical supply to the BACTROX must conform to all national and local electrical codes.

INSTALLATION (CONTINUED)

UV LIGHTING

Check if your laboratory or workspace contains sources of UV lighting. Sustained exposure to direct sunlight, UVC, or UV germicidal lighting around 254nm, will cause a rapid aging of BACTROX acrylic glass panels and arm port sleeves. Periodic use of long-wave (365nm) UV hand lamps for bacterial identification should not damage the acrylic glass. See the [Maintaining the Acrylic Glass Panels](#) entry on page 52 for more details.

LOW OXYGEN AMBIENT CONDITIONS

Low oxygen work area environments can be created by the operation of several hypoxia chambers or CO₂ incubators in an enclosed area with inadequate ventilation. Frequent door openings and leakages may lead to lowered oxygen levels as CO₂, N₂, and other laboratory gasses crowd out oxygen.

The highest level of O₂ concentration achievable by the BACTROX is equal to the ambient (room) concentration. A low oxygen workspace environment around the BACTROX may have the following adverse effects:

- It can hinder the ability of the BACTROX to meet a user selected O₂ set point.
- It can interfere with an O₂ sensor calibration when using an ambient air sample.

CONDENSATION MANAGEMENT

The BACTROX comes with an integral Peltier effect chiller for capturing excess humidity on an internal cold plate. The condensate is then collected in a small reservoir that drains through a plastic tube into the workspace chamber. An open container such as, a beaker or flask, must be placed beneath the drain tube and drained regularly.

INSTALLATION (CONTINUED)

LIFTING AND HANDLING

The BACTROX is heavy, and care should be taken to use appropriate lifting devices that are sufficiently rated for these loads. Follow these guidelines when lifting and handling the BACTROX workstation:

- Lift the BACTROX only from its bottom surface.
- Doors, handles, and knobs are not adequate for lifting or stabilization.
- Restrain the BACTROX completely while lifting or transporting so it cannot tip.
- Remove all moving parts, such as shelf spacers and trays, and secure all doors in the closed position during transfer to prevent shifting and damage.

Note: To prevent damage when moving the BACTROX, turn each of the four leveling feet completely clockwise.

LEVELING

The BACTROX must be level and stable for safe operation. Each BACTROX ships with four leveling feet. Insert one leveling foot into each of the four holes in the bottom corners of the workstation. Adjust the foot at each corner until the workstation stands level and solid without rocking. To raise a foot, turn it in a counterclockwise direction; to lower a foot, turn it in a clockwise direction.

INSTALLATION (CONTINUED)



Warning: Never introduce hydrogen in any quantity into the BACTROX. Hydrogen will destroy the oxygen-sensing zirconium dioxide sensor and the BACTROX's ability to regulate oxygen levels. Use of hydrogen in a BACTROX voids the warranty.

GAS SOURCES AND USAGE

The BACTROX achieves a hypoxic atmosphere (1 – 20% O₂ concentration) by using injections of nitrogen. These injections force oxygenated atmosphere out through a vent valve located on the back wall of the chamber, lower right side.

Before installing a BACTROX, contact your site safety officer and review your institutional safety protocols for handling, storing, securing, and using compressed gasses. Follow all local ordinances and national regulations regarding compressed gases in your research or production environment.

Note: Always use medical grade gas supply sources. Use of non-medical grade gases risk introducing contaminants into the chamber, may damage workstation components, and will void the unit's warranty.

Gas source considerations:

- A cylinder or wall source of medical grade nitrogen (N₂) is the primary injection gas for creating a hypoxic environment.
- For setting up a carbon dioxide-enriched chamber environment Sheldon Manufacturing recommends using a cylinder or wall source of medical grade carbon dioxide gas (CO₂) in addition to nitrogen.
- Always use a two-stage gas pressure regulator to ensure consistent metering for all gasses. Some single-stage regulators have two (2) gauges. Make certain your regulator is a two-stage regulator.
- The BACTROX uses injections of air drawn from the laboratory environment when required to raise the concentration of oxygen inside the workspace chamber.

Usage Rates

- The BACTROX requires approximately 400lbs of N₂ to purge the workspace chamber down to a 1% O₂ concentration.
- N₂ usage rates during normal operations vary considerably. Among the major factors driving usage are the O₂ set point (the lower the set point, the higher the rate of use) and if CO₂ is being added to the chamber atmosphere.
- Accessing and working in the workspace chamber significantly increases the consumption rate of N₂. Proper entry and exit techniques, along with proper movement technique while in the chamber, are essential for reducing N₂ usage. Proper techniques are covered in the Operation section of this manual (page 40).

INSTALLATION (CONTINUED)

INSTALL THE WORKSTATION

Install the unit in a workspace location that meets the criteria discussed in the previous entries of the Installation section.

Do not connect the unit to its power source at this time.

Note: Always match the correct type of gas regulator to the correct gas type. Example: A CO₂ regulator should always be used for CO₂ gas applications.

CONNECT TO GAS SOURCES

1. Install the dual stage gas regulators on the gas-source cylinders to be used for your application.
2. Set gas regulators for BACTROX supply at **15 – 20 PSI**.
3. Do not exceed 20 PSI!

PSI	Megapascals	Kilopascals	Bar
15 - 20 PSI	0.103 – 0.138 Mpa	103.42 – 137.89 Kpa	1.03 – 1.38 bar

4. Connect the nitrogen (N₂) gas regulator or wall source to the N₂ IN port.
 - a. The nitrogen and carbon dioxide regulators provided with the BACTROX each comes with ¼ inch OD tubing to connect to the BACTROX.
5. If using CO₂ in addition to nitrogen, connect the CO₂ regulator or wall source to the CO₂ IN port.
6. **Do not open** the gas valves at this time.
 - a. Gas will be supplied to the BACTROX when establishing a hypoxic atmosphere, as part of the Preparation for Use procedure in the Operation section.

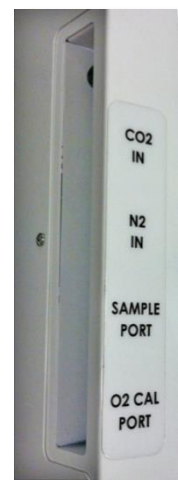


Figure 8: Gas Ports

INSTALLATION (CONTINUED)

INITIAL CLEANING

Disinfect the workstation to the standards of your laboratory or production environment protocol prior to placing the unit into operation. The BACTROX was disinfected at the factory prior to shipment. However, Sheldon Manufacturing cannot guarantee that the BACTROX was not exposed to contaminants en route, or that the factory procedure matches the standards of your institutional protocols. See the **Cleaning** procedure in the User Maintenance section on page 51 for more information about cleaning and disinfecting the BACTROX. Also see step 2 of the Install Components procedure below.

DEIONIZED AND DISTILLED WATER

Note: Do not use deionized water for cleaning or humidifying your BACTROX.

While DI water is useful in variety of laboratory applications, it is an aggressive solvent that attacks most metals. Use of DI water for cleaning or humidification in a Shel Lab incubator voids the unit warranty. Sheldon Manufacturing recommends the use of distilled water in the resistance range of 50K Ohm/cm to 1M Ohm/cm, or a conductivity range of 20.0 uS/cm to 1.0 uS/cm, for cleaning and humidifying applications.



Figure 9: Arm Port Doors

INSTALL ACCESSORIES

Place the following components in the workstation:

1. Remove all protective wrappings from accessories, shelving, and the workstation.
2. Clean, disinfect, and place the following items in the workspace chamber:
 - a. Arm port doors through pass box. The doors may be stored in the hanging slots on the bottom of the workspace incubator during the setup.
 - b. The calibration O₂ sensor cover plate through arm ports or pass box.
 - c. The incubator bottom shelf spacers through pass box.
 - d. The petri dish racks through arm ports. These can be placed on the top shelf of the workspace chamber incubator during the setup.
 - e. A glass flask or beaker placed under the plastic condensation drain tube on the left side of the chamber.
 - f. Any equipment or containers that will be used in the workspace chamber. Doing so now saves time and nitrogen by eliminating pass box purge cycles. If possible, containers should be placed in the chamber loose lidded or open to allow oxygen to be evacuated during hypoxic atmosphere setup. Sealed containers act as reservoirs of oxygen.



Figure 10: Incubator Shelf Spacers



Figure 11: Petri Dish Racks

INSTALLATION (CONTINUED)

SHELVING INSTALLATION

Install the three (3) metal bottom spacers included with the BACTROX on the bottom shelf of the workspace chamber incubator. These spacers shield sample containers from direct contact with the warm surfaces of the incubator, and ensure uniform heat dissipation.

1. Set the spacers on the bottom shelf of the workspace chamber incubator, side by side, with the “SPACER” label facing outwards toward you.
2. An empty plate or dish placed at the bottom of each sample stack can provide extra shielding for microaerophiles that are heat sensitive, or if the incubator is being run at high temperatures.
3. **Do not load samples at this time.**
4. Leave both doors of the workspace chamber incubator slightly open to prepare the unit for establishing a hypoxic atmosphere. Failure to leave the doors open during the setup will create reservoirs of ambient oxygen atmosphere. **Leave the doors approximately 1 cm (0.5 inches) open.** Opening the doors all the way at the start of the purge will cause the incubator to significantly overshoot its set point when the doors are finally closed



5680502

Figure 12:
Incubator Shelf
Spacers

INSTALL ARM PORT DOORS

Carryout the steps below to install the arm port doors in the ports. The doors should already be inside the workspace chamber.



1. Turn the silver locking bar to a roughly 45° position. 
2. Install each door bottom first, inserting the tabs into the slots on the arm ports. (See the [Exiting the Chamber procedure](#) on page 41 for pictures of this process.)
3. Tilt the door up so that it fits securely within the arm port.
4. Turn the locking bar to the horizontal position: 
5. Secure the door by turning the black arm port door knob clockwise, using wrist strength only, until the knob grabs and feels snug.
 - a. Tightening too much may compromise the integrity of the door by pulling out of position the post that the knob and locking bar are mounted on.



Figure 13: Arm
Port Doors with
wrappings

INSTALLATION (CONTINUED)

INSTALL THE SLEEVE ASSEMBLIES

Start with either side.

1. Unroll the large opening of a sleeve over the plastic lips of the arm port door opening.
2. Secure the sleeve to the arm port using the 48 inch (121cm) self-gripping strap included with the sleeve assembly.
3. Repeat the process for the 2nd sleeve assembly and arm port.

Note: Sleeve assemblies can be left attached to the BACTROX when not in use. No stowing procedure is required.



Figure 14: Mounted Sleeve Assemblies

4. Insert both sleeve plugs.



Figure 15: Inserting Sleeve Plugs

INSTALLATION (CONTINUED)

REMOVE PASS BOX VENT COVER

Remove the rubber cover from the pass box vent, located on the back of the BACTROX on the lower right corner.






Figure 16: Pass Box Vent

GRAPHIC SYMBOLS

The BACTROX is provided with multiple graphic symbols located on its exterior and interior surfaces. The symbols identify hazards and the functions of the adjustable components, as well as important notes found in the user manual.

Symbol	Definition
	Indicates that you should consult your user manual for further instructions. Indique que l'opérateur doit consulter le manuel d'utilisation pour y trouver les instructions complémentaires.
	Indicates Temperature Repère température
	Indicates the Over Temperature Limit system Indique le système de dépassement de température
	Indicates AC Power Repère le courant alternative
	Indicates I/ON and O/OFF I repère de la position MARCHÉ de l'interrupteur d'alimentation O repère de la position ARRÊT de l'interrupteur d'alimentation
	Indicates protective earth ground Repère terre électrique
	Indicates UP and DOWN respectively Touches de déplacements respectifs vers le HAUT et le BA
	Indicates Manually Adjustable Indique un bouton réglable manuellement
	Indicates Potential Shock Hazard Signale danger électrique

GRAPHIC SYMBOLS (CONTINUED)

Symbol	Definition
	WEEE Directive compliant logo Indicates the unit should be recycled (Not disposed of in land-fill) Indique l'appareil doit être recyclé (Ne pas jeter dans une décharge)
	Indicates an injection of gas Indique un flux de gazeux
	Indicates the incubator heater is active L'élément chauffant est la production de chaleur

CONTROL PANEL OVERVIEW



Figure 17: Control Panel



Power Switch

The power switch controls all power to the BACTROX and its systems. The switch illuminates when in the ON (I) position, along with all three of the green digital displays



Figure 18:
Temperature &
OTL Controls

Main Temperature Control and Green Digital Display



Marked SET TEMPERATURE °C, this display shows the current incubator air temperature accurate to within $\pm 0.1^\circ\text{C}$. The arrow buttons can be used to adjust the temperature set point, mute a temperature deviation alarm, or place the unit in its calibration mode and enter a display value correction.



The green pilot light located beneath the label HEATING ACTIVATED illuminates whenever the workspace incubator heating elements are powered and warming the incubator.

Set Over Temperature



This graduated dial sets the temperature limit for the Over Temperature Limit system. The OTL System prevents unchecked heating of the incubator in the event of a failure of the main temperature controller. For more details, please see the explanation of the [Over Temperature Limit System](#) on page 30 in the Operation section Theory of Operation.



Marked OVER TEMP ACTIVATED, the light illuminates when the OTL System is routing power away from the incubator heating elements.

CONTROL PANEL OVERVIEW (CONTINUED)



Figure 19: O₂ and CO₂ Controls

Set OXYGEN %

The SET OXYGEN % display shows the oxygen concentration in the workspace chamber as a percentage of the chamber atmosphere between 0 and 25%. The control **UP / DOWN** arrow pad is used to input an O₂ concentration set point for the workspace chamber atmosphere between 0.5 and 20%, and to enter calibration offsets. The display is also provided with LED deviation alarms and an audible buzzer alarm. These alarms activate in the event of a deviation in O₂ concentration of $\pm 1\%$ from the O₂ set point. The yellow LED marked MUTE illuminates whenever an audible O₂ deviation alarm is being muted. There is a 15 minute delay for the activation of the low deviation buzzer alarm.



N2 Injecting / Air Injecting

The green N2 pilot lights illuminates when unit is injecting nitrogen drawn from the N2 IN port into the workspace chamber. The Air Injecting light activates when outside air is being injected to raise the oxygen concentration of the chamber to match the user-selected O₂ set point. The N2 solenoid operation is normally accompanied by a clicking sound. The air pump creates a slight buzzing.



SET CO₂%

Labeled SET CO₂, this panel includes a green digital LED display connected to an infrared sensor probe. The probe measures the concentration of CO₂ in the workspace chamber as a percentage of the chamber's atmosphere. The control's **UP / DOWN** arrow pad is used to input a CO₂ concentration set point for the workspace chamber atmosphere, and to enter calibration offsets. The display is also provided with LED deviation alarms and an audible buzzer alarm. These alarms activate in the event of a deviation in CO₂ concentration of $\pm 1\%$ from the CO₂ set point. The yellow LED marked MUTE illuminates whenever an audible CO₂ deviation alarm is being muted. There is a 15 minute delay for the activation of the low deviation buzzer alarm.



CO₂ Injecting Light

Marked CO₂ INJECTING, this pilot light will illuminate whenever the unit's CO₂ solenoid is injecting CO₂ into the workspace chamber from the CO₂ In port. CO₂ solenoid operation is normally accompanied by a clicking sound.



CONTROL PANEL OVERVIEW (CONTINUED)



Figure 20: Bass Box Switch and Chamber Light



Start Pass Box Purge

The START PASS BOX PURGE button initiates an approximately 45 second long cycle of the pass box chamber. The cycle purges oxygenated atmosphere by injecting nitrogen into the box chamber and forcing oxygenated atmosphere out through a vent on the back of the unit.



Pass Box Purging

This clear light illuminates for the duration of a pass box purge cycle.



Lights

The LIGHTS switch turns the workspace chamber lights on and off.

Fuse

Located on the inside the power cord inlet on the back of the workstation, the fuse protects against over current conditions. If the fuse blows, the BACTROX will shut down. The cause of a blown fuse should be determined prior to replacing it.

THEORY OF OPERATION

The BACTROX is engineered to generate low-oxygen (hypoxic) atmospheres in its workspace chamber. Where normal sea level atmosphere typically has an oxygen concentration of around 20.7%, the BACTROX can reliably achieve a range of O₂ concentrations from 0.5 – 20%, with the high end of the operating range dependent on the available ambient O₂. Additionally, the chamber atmosphere can be enriched with a CO₂ concentration of 1 – 20% when connected to a CO₂ source. The unit is equipped with arm ports and an airlock-style pass box, which allow samples and equipment to be introduced to, manipulated in, and removed from the chamber without compromising the hypoxic atmosphere. The workstation is provided with a cabinet-style incubator with a range of 60°C down to the ambient room temperature + 8°C for culturing or cultivation applications.

Achieving and Maintaining Hypoxic Conditions

The BACTROX achieves low-oxygen conditions through injections of non-oxygen gases. These force oxygenated atmosphere out through a vent located on the chamber back wall. Nitrogen (N₂) is the primary gas for such injections, along with any CO₂ enrichment injections. In the event O₂ levels fall below the user-programmed set point, the BACTROX injects oxygenated atmosphere drawn from the surrounding laboratory air.

Maintenance of the O₂ and CO₂ set points is a dynamic competition of N₂ and air injections, as well as CO₂ injections. When the workstation is sitting sealed and undisturbed N₂ injections will take place approximately every 4 – 6 seconds. The N₂ and CO₂ gas solenoids produce audible clicking sounds when injecting. The air pump produces an audible buzz when injecting ambient atmosphere.

Pass box operations or a user working with arms in the chamber will displace some hypoxic atmosphere, and result in increased N₂ and CO₂ usage. Proper arm port entry and exit techniques, minimized pass box usage, and slow, deliberate movements while working in the chamber can significantly reduce the displacement of hypoxic atmosphere.

The BACTROX microprocessor controller employs proportional-integral-derivative analytical feedback-loop functions when measuring and controlling gas levels. PID-controlled gas injection lengths are proportional to the difference between the measured chamber gas concentration and the current set points. The frequency of injections are derived from the rate of change in the difference. Gas injections slow as the chamber atmosphere nears the set points to avoid overshoots.

OPERATION (CONTINUED)

Sensing Oxygen

The BACTROX detects the concentration of oxygen in the chamber atmosphere using the black and silver O₂ sensor located on the left chamber wall. The O₂ sensor operates with an internal temperature of 700°C to produce mobile oxygen ions through disassociation. The volume heated to temperature is very small. The ions are drawn into sensor cavities separated by porous sheets of platinum-coated zirconium dioxide that act as an electrolyte. The controller determines the partial pressure of oxygen by measuring the voltage potential differential between ions separated by the zirconium sheets. That measurement is then compared with reference differentials that were saved in the controller memory during the previous O₂ sensor calibration. The controller derives the O₂ concentration from the measured partial pressure of oxygen (PPO).

The sensor must be allowed five minutes to warm up to its operating temperature in order to provide an accurate O₂ measurement.

Each BACTROX is shipped with the sensor calibrated for a two point curve using an N₂ – O₂ reference mixture of 5% O₂ – 95% N₂ for the low end. An ambient air sample with a calculated concentration was used for the high end.

The O₂ sensor comes set with an output range of 0 – 25% O₂ concentration. Control panel display readings above 25% are indicative that the sensor and O₂ display are out of calibration. The unit controller has an effective operating range of 0.5 – 20%. Sheldon Manufacturing cannot guarantee a stable oxygen concentration at settings above 20%. In part due to the variability of ambient oxygen to draw on, as well as significantly increased wear on the air motor pump when running near continually to maintain an ambient atmosphere in a sealed chamber containing metabolically active samples.

Sensing Carbon Dioxide

The BACTROX monitors CO₂ levels with an infrared sensor. The sensor operates on the principle that a band of infrared light is absorbed by CO₂. The higher the CO₂ concentration in the chamber atmosphere, the more of that band of infrared is absorbed. This means that sensor is only sensitive to CO₂, so measurement accuracy is consistent, regardless of the presence of other gasses in the incubator. The addition of CO₂ to the chamber atmosphere is purely optional and dependent on your laboratory protocol and application requirements.

OPERATION (CONTINUED)

Accessing the Workstation

Items such as media containers and laboratory equipment can be introduced to or removed from the hypoxic workspace through the pass box. The pass box runs a user-initiated 45-second nitrogen purge that forces oxygenated room atmosphere out of the pass box chamber through a valve on the back of the unit.

Users can access and work glove-free in the workspace chamber by donning the sleeve assemblies attached to the front panel arm ports, and entering through the ports. Effective use of the sleeve assemblies requires bare skin contact between the widest part of the user's forearms and the cuff ring of the sleeve assembly. Smooth, small items held in hand may be introduced into the workspace chamber through the sleeve assemblies.

The sleeve assemblies are compatible with exam gloves for handling pathogenic samples inside the workspace.

Incubator

The BACTROX microprocessor controller monitors and controls the incubator temperature using a solid state thermometer probe attached to the incubator body, along with two heating elements. As with gas injections, heating is monitored and controlled by the microprocessor controller using PID feedback loops. PID-controlled heating is proportional to the difference between the measured incubator temperature and the user-programmed temperature set point. The rate of heating pulses is derived from the rate of change in the difference. The integrator function slows the rate of heating slows as the measured chamber temperature nears the set point to avoid overshooting.

The BACTROX incubator relies on natural heat radiation for cooling.

The PID analytics allow the controller to optimize heating rates for the local temperature conditions. If the BACTROX is moved to a new location with a significant temperature difference from its previous surroundings, it may require 24 hours of incubator run time for the controller to fully adapt to the new thermal environment. For example, a BACTROX that has optimized to use more power for heating in a cool environment may generate overshoots for a day after being moved to a warmer location.

This is why the incubator should be run at its application set point for 24 hours prior to performing a temperature calibration if the BACTROX has been moved to a new location. Additionally, accelerated heat loss from leaving the incubator doors open for long periods (an hour or more) can trick the controller into behaving as though it is operating in a cool environment.

Optimization helps maintain a high degree of temperature stability and reduce overall power usage during normal operating conditions.

OPERATION (CONTINUED).

The Over Temperature Limit System (OTL)

When set, the OTL system prevents runaway heating in the event of a failure of the microprocessor controller board or its thermometer probe. It does so by depowering the heating elements whenever the temperature in the incubation chamber exceeds the OTL setting. Typically the OTL is set approximately 1°C above the incubator set point. Because of its nature as a mechanical cutoff system and its lack of PID analytics, the OTL cannot deliver the same degree of temperature stability and measurement precision as the digital display and microprocessor board. The OTL System should only be used as a means of heating regulation for the incubator until a failed controller board or its thermometer probe can be repaired or replaced.

Com Port Data and Logging Outputs

The BACTROX comes with a RS232 data port, and generates three outputs once per minute as a logline describing the oxygen, CO₂, and temperature levels. These outputs can be logged using a computer or building management system connected to the RS232 port. Please see the [Data Output and Logging](#) entry on page 82 for a more detailed description.

Condensation Management

Petri plates and other open or breathable containers loaded with sample can generate significant humidity and condensation in the workspace chamber if unchecked. To control humidity levels the BACTROX is provided with a Peltier effect condensate chiller located behind a circulation fan on the upper left side of the workspace chamber. Chamber humidity is trapped on the chiller's cold plate in the form of condensation. The condensate is then channeled into a drain tube that empties into a receptacle placed in the workspace chamber by the end-user. The receptacle must be regularly drained. The condensate chiller and its circulation fan eliminate the need to use chemical desiccants to control humidity level. Desiccants can dry out culture media while leaving significant condensate untouched. Under normal operating conditions the chiller can handle media evaporation from up to 300 plates.

The condensate chiller is active whenever the BACTROX is powered.

OPERATION (CONTINUED).

Note: Preparing the BACTROX for use requires approximately three hours of work. However, Sheldon Manufacturing recommends allowing 24 hours for the chamber incubator to heat and stabilize and for the hypoxic atmosphere to stabilize, prior to loading samples in the workstation.

PREPARING THE BACTROX

Verify that the following procedures were completed during the workstation installation:

- [Connect to the Gas Supply](#) page 17
- [Install Components](#) page 18
- [Shelving Installation](#) page 19
- [Install the Arm Port Doors](#) page 19
- [Install the Sleeve Assemblies](#) page 19

Perform the following procedures to prepare the unit for use:

- ✓ [Power the BACTROX](#) page 32
- ✓ [Verify O₂ Sensor Accuracy](#) (Optional) page 33
- ✓ [Establish a Hypoxic Atmosphere](#) page 36
- ✓ [Setting the Incubator Temperature](#) page 39
- ✓ Read [Chamber Entry](#) page 40
- ✓ Read [Chamber Movement Techniques](#) page 40
- ✓ Read [Exit the Chamber](#) page 41
- ✓ Read [Operating the Pass Box](#) page 42
- ✓ [Set the Over Temperature Limit](#) page 43
- ✓ [Load the Workstation](#) page 44

Note: **Audible gas and temperature deviation buzzer alarms may sound** during the setup while the BACTROX is establishing a hypoxic chamber atmosphere and the incubator is warming up. **To mute these alarms**, press the up or down arrow on each control that a red deviation indicator is illuminated on. Hold the button until the yellow MUTE indicator illuminates. All active alarms must be muted before the buzzer will shut off. There is a fifteen minute delay for the activation of audible low deviation alarms.

OPERATION (CONTINUED).

POWER THE BACTROX

Note: Before plugging in the BACTROX, verify that the wall power supply and workstation electrical requirements have been checked for compatibility (see the Installation section, page 13).



1. Plug the female end of the power cord into the inlet on the back of the workstation.
2. Plug the male end of the power cord into the earth-grounded outlet.
3. Turn the power switch ON (I) to verify that the workstation will power up.
4. The following lights and displays should illuminate. If they do not, consult Technical Support:
 - a. The Green power switch.
 - b. All three green digital displays on the main control panel.
5. The three displays will initially show fluctuating gas and temperature levels. It will take approximately five minutes for the gas sensor probes come up to operating temperature.

OPERATION (CONTINUED)

VERIFY O₂ SENSOR ACCURACY








Optional: The BACTROX is shipped with the oxygen sensor factory calibrated on a two-point curve at approximately 20.7% and at 5% at near sea level. An onsite verification of the calibrated sensor accuracy can be performed prior to establishing a hypoxic atmosphere, if required by your laboratory, study, or production protocol. **Sheldon Manufacturing strongly recommends performing a verification if you are preparing your BACTROX for use at altitudes above 2000 feet (600 meters).** Altitude-related pressure differences can result in offset errors of 0.1% or greater in display output.

A fast verification can be performed using one of two methods:

- **Reference Gas Verification.** Attach a bottle of calibration reference gas to the O₂ CAL PORT. After 5 minutes of inflow verify that the O₂ display matches the oxygen concentration of the calibration gas.
 - See next page for procedure.
- **Atmosphere sample.** Open the arm port doors and pass box, calculate the current ambient O₂ concentration. Verify that the O₂ display matches the ambient concentration, after 5 minutes of run time.
 - See page 35 for procedure.

Set O₂ to OFF

Before using either method, set the O₂ display to Off. This preserves the tuning of the controller's PID analytic feedback loops, and prevents the BACTROX from attempting to continually inject nitrogen during the procedure.

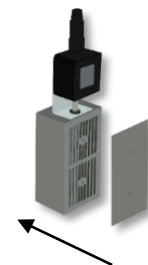
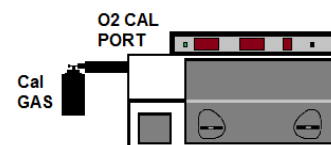
Set O ₂ to Off		
 OR 	<p>1. Press either one of the O₂ control arrow keys and then release.</p> <p>a. The display will briefly flash the letters "SP", then switch from bright to dim, and blink on and off.</p>	<p>SET O₂%</p> 
  Wait 5 Seconds	<p>2. Press and hold the down arrow key.</p> <p>a. Hold the key until the displayed percentage reaches 0, and then switches to "OFF".</p> <p>b. After you have adjusted the display to Off, wait five seconds.</p> <p>c. The display will cease flashing, brighten, and show the ambient level of O₂ in the chamber. The control is now set to Off, and the BACTROX-2 will not attempt to inject N₂.</p>	<p>SET O₂%</p>  <p>SET CO₂%</p> 

OPERATION (CONTINUED)

Note: Allow the BACTROX to run for at least 5 minutes prior to conducting the Verify O₂ Sensor Accuracy procedure. This is the time required for the sensor to heat up to its operational temperature and output an accurate O₂ concentration reading.

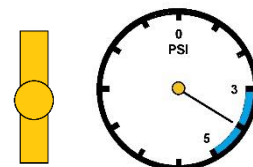
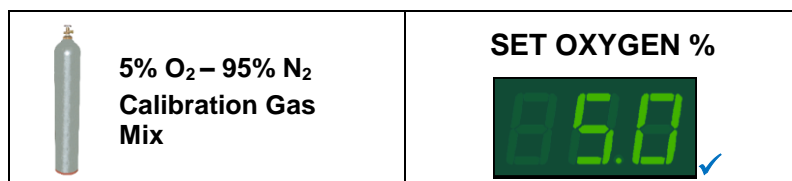
Verification Using Calibration Reference Gas Sample

1. Set the O₂ set point to OFF. This preserves the tuning of the controllers PID analytics, and prevents the unit from continually attempting to inject N₂
2. Attach the calibration gas cylinder to the BACTROX O₂ Cal Port.
3. Attach the calibration kit sensor cover to the O₂ sensor in the workspace chamber.
4. Set the calibration gas cylinder regulator to between **2 and 3 psi**.
5. Start a gas flow from the supply cylinder to the sealed O₂ sensor space.
6. Wait 5 minutes to achieve a sufficient concentration of calibration gas in the O₂ sensor space
7. Verify that the Set Oxygen % display matches the O₂ concentration of the calibration gas.



Wait 5 Minutes

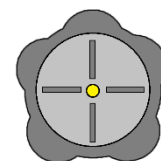
Example



Calibration Gas Regulator

8. Set the calibration gas regulator to closed; disconnect the cylinder from the BACTROX-2; reattach the O₂ Cal port cover; and remove the calibration kit cover from the O₂ sensor before continuing the Setup.

If the verification does not produce an acceptable match between the calibration gas sample and the BACTROX Set Oxygen % display, continue and complete the Preparing the BACTROX process. A full calibration procedure can be performed after to correct any errors in the accuracy of measurement or display output. Please see the [O₂ Sensor Calibration Procedure](#) on page 54 in the User Maintenance section.



Start Gas Flow

Note: The BACTROX O₂ display will be turned back on and an O₂ set point set during the [Establish a Hypoxic Atmosphere procedure](#) on page 36.

OPERATION (CONTINUED)

Note: Allow the BACTROX to run for at least 5 minutes prior to conducting the Verify O₂ Sensor Accuracy procedure. This is the time required for the sensor to heat up to its operational temperature and output an accurate O₂ concentration reading.


Verification Using Ambient Atmosphere Sample



Wait 5 Minutes

1. Open the arm ports and both pass box doors to expose the chamber to the room atmosphere.
2. Wait at least five minutes to ensure that the O₂ level of the chamber matches that of the ambient atmosphere.
3. Calculate the concentration of O₂ in the ambient atmosphere. See the [Calculating the O₂ Concentration Appendix](#) on page 84 for how to derive the current O₂ concentration of the ambient air.
4. Verify that your calculated ambient concentration matches the concentration shown on the Set Oxygen % Display.

Example

<ol style="list-style-type: none">1. $WVP = \left(\frac{H_{rel}}{100}\right) \cdot WVP_{max}$2. $PPO_2 = (BP - WVP)$3. $O_2\% = \left(\frac{PPO_2}{BP}\right) 100$4. = 20.7% O₂	<p>SET OXYGEN %</p> 
---	---

5. Reseal the arm ports, close and latch both arm port doors.

If the verification does not produce an acceptable match between the calculated value of the ambient sample and the Set Oxygen % display, continue and complete the Preparing the BACTROX process. A full calibration procedure can be performed after to correct any offset errors in the accuracy of measurement or display output. Please see the [O₂ Sensor Calibration Procedure](#) on page 54 in the User Maintenance section.

Note: The BACTROX O₂ display will be turned back on and an O₂ set point set during the [Establish a Hypoxic Atmosphere procedure](#) on page 36.

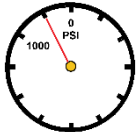

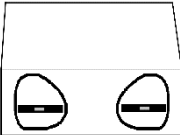
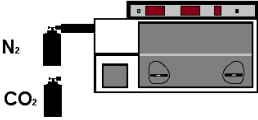
End of procedure

OPERATION (CONTINUED)

ESTABLISH A HYPOXIC ATMOSPHERE

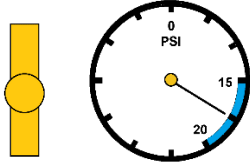
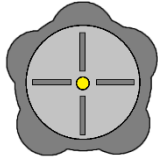
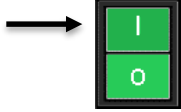





Carry out the following steps to purge the ambient atmosphere from the workstation and establish a hypoxic environment.

This procedure requires 1 to 3 hours to complete depending on experiencing at using the BACTROX, and the gas concentrations to be achieved. Under normal conditions, 1.5 to 2 hours are required to achieve an O₂ concentration of 1%. It may take up to 4.5 hours to achieve a 5% CO₂ concentration in the chamber. Normally **400lbs of N₂ tank pressure are required for a purge**, though this will be reduced if CO₂ is also being introduced to establish a carbon dioxide-enriched environment.

Establish Hypoxic Atmosphere	
<ol style="list-style-type: none"> Record the supply readings on the gauges of the gas sources. This is done to start a record of gas usage. 	 <p>Supply Reading</p>
<ol style="list-style-type: none"> Verify that both incubator doors are slightly open to avoid leaving a reservoir of fully aerobic atmosphere. See step 4 of the Shelving Installation Procedure on page 19. 	
<ol style="list-style-type: none"> Verify the arm port doors are closed and sealed. 	 <p>Arm Ports Secured</p>
<ol style="list-style-type: none"> Verify that the gas supply(s) is connected to the BACTROX. <ol style="list-style-type: none"> See the Connecting to the Gas Sources procedure on page 17 in the Installation section. 	








Procedure continued

OPERATION (CONTINUED)

Establish Hypoxic Atmosphere (Continued)	
<p>5. If not already done, set the N₂ regulator or source control to flow between 15 and 20 PSI.</p> <ol style="list-style-type: none"> Set the CO₂ regulator or source control to flow 15 to 20 psi if you will be setting up a CO₂ environment. Do not exceed 20 PSI! 	 <p>N₂ CO₂</p>
<p>6. Open the gas supply valves all the way to supply nitrogen to the BACTROX.</p> <ol style="list-style-type: none"> Do the same for the CO₂ regulator or in-house source, if you will be setting up a CO₂-enriched environment. 	 <p>N₂ CO₂</p>
<p>7. Turn on the BACTROX, if it is not already powered up.</p>	
<p>Note: If you have previously turned off the O₂ display as part of the optional Verification procedure on page 33, completing this step will turn the display back on.</p> <p>8. Set the oxygen set point to your application set point using the Up and Down arrows of the SET OXYGEN % control.</p> <ol style="list-style-type: none"> Press either of the arrow keys and then release. The display will briefly flash the letters “SP” for set point, then switch from bright to dim and blink on and off. This is the adjustable set point. Use the up and down keys to adjust the set point to your required O₂ concentration. After adjusting the display to your application or process set point, wait 5 seconds. The display will cease flashing, and brighten. The set point has now been entered into the BACTROX, and the workstation will now begin injecting N₂ to create a hypoxic atmosphere. <div style="display: flex; align-items: center;">  <p style="margin: 0 5px;">OR</p>  </div> <div style="display: flex; align-items: center; margin-top: 10px;">  <p style="margin-left: 5px;">Wait 5 Seconds</p> </div>	<p>SET OXYGEN %</p>  <p>SET OXYGEN %</p>  <p>Note: 15% is an arbitrary example value.</p>

Procedure continued on next page

OPERATION (CONTINUED)

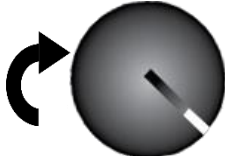









Establish Hypoxic Atmosphere (Continued)	
<p>9. If you will be using a carbon dioxide-enriched atmosphere, set the CO₂ set point using the up and down arrows of the SET CO₂ % control .</p> <div style="display: flex; align-items: center;">  <p style="margin: 0 5px;">OR</p>  </div> <p style="margin-top: 10px;">  Wait 5 Seconds </p> <ol style="list-style-type: none"> a. Press either of the arrow on the keys and then release. b. The display will briefly flash the letters “SP”, then switch from bright to dim and blink on and off. c. Use the up and down keys to adjust the set point to the CO₂ concentration you will be using for your application. d. After adjusting the display to your application or process set point, wait five seconds. The display will cease flashing, and brighten. The set point has now been entered in to the BACTROX, and the work station will now begin injecting CO₂ to establish a carbon dioxide-enriched atmosphere. 	<p style="text-align: center;">SET CO₂ %</p> <div style="text-align: center;">  </div> <p style="text-align: center;">SET CO₂ %</p> <div style="text-align: center;">  </div> <p style="text-align: center;"><i>Note: 5% is an arbitrary example value.</i></p>
<p>10. Do not enter the workspace chamber or use the pass box while the BACTROX is purging the chamber atmosphere.</p>	<div style="text-align: center;">  </div>
<p>11. Set the incubator temperature set point (optional)</p> <ol style="list-style-type: none"> a. While waiting for the oxygen purge to complete, you may set the temperature set point of the workspace incubator to your application’s required temperature. b. Please see the Setting the Incubator Temperature procedure on the next page. 	<p style="text-align: center;">Optional Setting the Incubator Temperature Page 39</p>
<p>12. End states. A hypoxic atmosphere has been achieved when:</p> <ol style="list-style-type: none"> a. The SET OXYGEN % display has stabilized for 1 hour without any changes at or near your O₂ concentration set point. Under normal conditions 1.5 to 2 hours are required to drop down to a concentration of 1% from ambient. b. ...the SET CO₂ % display has stabilized without any changes for 1 hour at or near the correct CO₂ concentration. Under normal conditions 45 minutes are required to go up to a 1% concentration. It may take significantly longer to reach a final CO₂ concentration than the O₂ concentration. 	<div style="text-align: center;">  </div>

End of procedure

OPERATION (CONTINUED)

SET THE INCUBATOR TEMPERATURE

Adjust the incubator temperature set point to that of your application.

Set Temperature	
<ol style="list-style-type: none"> Turn the Over Temperature Limit control dial clockwise to the maximum position indicated by the largest dot, if it is not already set to max. <ol style="list-style-type: none"> This prevents the Over Temperature Limit system from interfering with the Set the Temperature Set Point procedure. 	
 <p>OR</p>  <ol style="list-style-type: none"> Press either the Up or Down key on the Set Temperature control panel to activate the temperature set point mode. <ol style="list-style-type: none"> The temperature display will briefly flash the letters “SP” to indicate a Set Point is about to be displayed. The Set Temperature digital display will then show the flashing adjustable temperature set point. 	<p>Set Temperature °C</p> 
 <p>OR</p>  <ol style="list-style-type: none"> Use the Up or the Down arrow key to adjust the set point. <ol style="list-style-type: none"> If neither key is pressed within 5 seconds, the Set Temperature display will stop flashing. It will then return to displaying the current air temperature of the incubator chamber. 	<p>Set Temperature °C</p> 
 <p>Wait 5 Seconds</p> <ol style="list-style-type: none"> Wait 5 seconds after entering your set point. <ol style="list-style-type: none"> The display will stop flashing, and the set point is now saved in the controller. The incubator will now automatically heat or passively cool to match your set point. The display will revert to showing the current chamber air temperature. 	<p>Set Temperature °C</p> 
<ol style="list-style-type: none"> Wait 24 hours for the incubator temperature to stabilize, then set the Over Temperature Limit system. See page 43. 	

End of Procedure


Note: Sheldon manufacturing recommends waiting 24 hours for the incubator temperature to stabilize prior to loading samples.

OPERATION (CONTINUED)

Note: Familiarize yourself with the chamber entry procedure and movement techniques prior to accessing a hypoxic chamber.

CHAMBER ENTRY

Perform these steps to access the workspace chamber without drawing in ambient atmosphere from outside the BACTROX. See the [Exiting the Chamber procedure](#) on page 41 for how to withdraw your arms from the chamber and sleeve assembly without compromising the hypoxic atmosphere.

1. Remove watches, bracelets, large rings, and any sharp objects that might damage the cuffs or sleeves.
2. Remove the sleeve plugs if installed.
3. Place your hands in the sleeve assemblies so that the cuff rings are secure around the bare skin of the widest part of your forearms.
 - a. The BACTROX comes with size 9 cuffs. Size 6 cuffs are available for users with smaller forearms; size 8 for midsized users. Please see the [Parts List](#) on page located on page 80.
4. Loosen both arm port door knobs by two or three turns if installed
5. Rotate the locking bars to a roughly 45° diagonal: 
6. Slowly push one door into the chamber, then the second. The Arm port doors can be hung in the arm port door holders on the bottom of the workspace chamber incubator.

CHAMBER MOVEMENT TECHNIQUES

The BACTROX workspace chamber is a mostly sealed environment, with a filtered vent for exhaust and pressure relief. Entering through the arm ports and reaching into the chamber will create pressure from the displacement of atmospheric volume by the solid mass of your arm. This generates resistance while working in the confines of the chamber. Entering and moving in the chamber will also force some of the hypoxic atmosphere out of the chamber, and draw in a corresponding amount of the aerobic ambient atmosphere. The entry of outside atmosphere will in turn increase gas usage.

Use the following techniques to manage pressure and reduce gas use.

1. Avoid making fast or large movements while working in the chamber.
2. Use a swimming motion, withdrawing one arm partly into the arm port while reaching in with the other. Placing both arms in the chamber at the same time displaces a larger total volume of hypoxic atmosphere.

OPERATION (CONTINUED)

Note: Familiarize yourself with the chamber exit procedure prior to accessing a hypoxic chamber.

EXIT THE CHAMBER

The following steps are used to exit the chamber without pulling in ambient atmosphere.

1. Check that the interior pass box door is closed and secured to avoid drawing oxygenated atmosphere through the pass box.
2. Remove the arm port doors one at a time from the hanging stowage slots beneath the incubator.
3. Place each door on the chamber floor by its corresponding port.
4. Grasp one door by the silver locking bar, and **slowly** withdraw your arm holding the door. Inset the tabs on the door into the slots of the arm port assembly. Then tilt the door toward you so that it sits securely in the port. Rotate the locking bar from the diagonal to horizontal position.
5. Tighten the arm port door knob until it starts to grab, using wrist strength only.
6. Repeat steps 4 – 5 for the second door.
7. Withdraw one hand from its cuff and exit the sleeve.
8. Withdraw your other hand from the second sleeve.
9. Optional: Install the sleeve plugs.

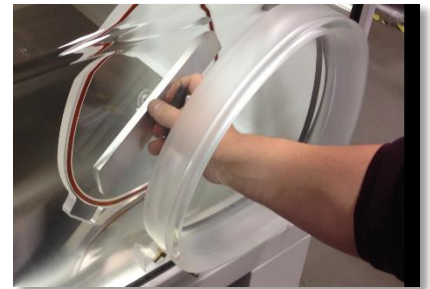


Figure 21: Securing the Arm Port Doors

OPERATION (CONTINUED)

OPERATING THE PASS BOX

Introducing Items

1. Make sure the inner pass box door is closed and secured.
2. Open the outer pass box door.
3. Load the pass box sliding shelf with items to be introduced in the workspace chamber.
4. Close and secure the outer door.
5. Press and immediately release **Start Pass Box Purge switch**. Do not hold down the switch.
6. The purge cycle runs for 45 seconds.
 - a. N₂ is injected into the pass box, and oxygenated atmosphere evacuated through a vent on the back of the BACTROX.
 - b. The clear PASS BOX PURGING light will flash throughout the cycle.
7. Once the purge cycle is complete you may open in the inner pass box door and pull the sliding shelf into the workspace chamber.



Figure 22: Pass Box

Best Practice. Stow the sliding shelf in the pass box, and close the inner pass box door once the shelf has been unloaded. This protects accidental exposure of the workspace chamber to fully aerobic atmosphere if the outer pass box door is opened while the inner door is still open.

Aborting: To abort a pass box purge cycle, press and hold the **START PASS BOX PURGE switch** for **five (5) seconds**. The purging light will cease flashing and turn off. The cycle is now terminated.

Removing Items

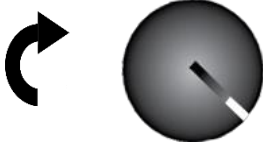

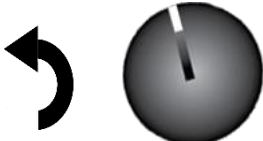

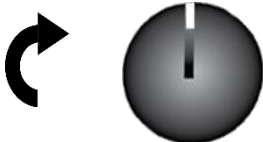

1. Make sure the outer pass box door is closed and secured.
2. Open the inner pass box door.
3. Pull the pass box sliding shelf into the chamber and load with items to be removed.
4. Return the shelf to the pass box. Close and secure the inner door.
5. Press and immediately release START PASS BOX PURGE switch. Do not hold down the switch.
6. The purge cycle runs for 45 seconds.
 - a. The clear PASS BOX PURGING light will flash throughout the cycle.
7. Once the purge cycle is complete you may open in the outer pass box door and unload the pass box shelf.

OPERATION (CONTINUED)

SET THE OVER TEMPERATURE LIMIT

This procedure sets the Over Temperature Limit to approximately 1°C above the current temperature set point. Wait at least 1 hour after changing the temperature set point. When setting up the BACTROX for use it is best to wait at least 8 hours after powering the unit. 24-hours is ideal.

Perform the steps below once the incubator has stabilized at the application set point temperature.

		Example
1. If you have not done so already, turn the Set Over Temperature Limit control dial clockwise to the maximum position.		
2. Turn the Over Temperature Limit control dial counterclockwise until the red Over Temp Limit Activated light illuminates.		
3. Slowly turn the dial clockwise until the Over Temperature Limit Activated light turns off. Stop turning the control.		
4. Leave the OTL dial set slightly above the activation point. The Over Temperature Limit is now set approximately 1°C above the current temperature set point.		

The Over Temperature Limit System activates if the incubator temperature exceeds the setting of the OTL System. If the BACTROX main controller has failed, or the OTL is set below your chosen temperature set point, the OTL alarm indicator will turn on and off indefinitely as the OTL depowers the heating elements. If this is happening, repeat steps 1 -3 to verify that the OTL is set **above** the temperature set point. If it is, contact [Sheldon Technical Support](#) for assistance, see page 6.

End of procedure

OPERATION (CONTINUED)

LOAD THE WORKSTATION

Sheldon Manufacturing recommends waiting 24 hours after establishing a hypoxic atmosphere and entering an initial incubator temperature set point before loading samples into the unit.

See page 42 for how to operate the pass box when introducing and removing items from the chamber.

The pass box sliding shelf can hold and transport up to 252 plates. Place samples and their media containers on the shelves inside the incubator as evenly spaced as possible. Good spacing allows for atmosphere circulation and a higher degree of temperature uniformity. If heat-sensitive microorganisms are being cultivated it may be necessary to place an empty petri plate at the bottom of each stack.

This concludes the Preparing the BACTROX portion of the Operation section

GAS CONSERVATION METHODS

1. Minimize the number of times per day the pass box is purged and opened.
2. Introduce small individual items such as, sealed micro plates or transport tubes, into the workspace chamber through the sleeve assemblies.

ACTIVATED CHARCOAL SCRUBBER AND REJUVENATION CYCLE

An activated charcoal scrubber may be placed in the workspace chamber to absorb volatile fatty acids (VFAs) or volatile sulfur compounds (VSCs) produced by sample cultivation. This helps keep the workstation interior clean and reduce odors when running cultivation processes or applications that produce large amounts of VFAs or VSCs. See the [Accessories section](#) on page 82 for Sheldon Manufacturing's recommended charcoal scrubber and Anatox fan scrubber unit.

1. The recommended use is 250 grams (one packet) placed in a 500ml beaker inside the chamber. Place another 250 grams into a second 500ml beaker.
2. On day two (2) replace the first beaker of charcoal scrubber with the second.
3. On day three (3) reactivate the first beaker of charcoal scrubber by heating at a minimum of 160°C for at least two (2) hours). Place the reactivated scrubber in the chamber. Remove and reactivate the second scrubber. For best effect, reactivate the scrubber by heating overnight.
4. Repeat this cycle for six (6) months. Discard both scrubbers after six months and replace.

OPERATION (CONTINUED)

ATTACHING EQUIPMENT TO THE CHAMBER ACCESSORY OUTLETS

BACTROX workstations are provided with two 1 amp accessory outlets located inside the workspace chamber, on the left wall. The power switch on the main control panel controls power to the accessory outlets. The outlets can power equipment such as magnetic stirrers or an Anatox activated charcoal scrubber fan. Do not attach equipment drawing more than one (1) amp.

Accessory equipment may produce additional heat in the workspace chamber. This can affect the temperature range of the incubator. Monitor the incubator performance when using powered accessories inside the workspace chamber.

HUMIDIFYING THE INCUBATOR

Placing only a small number of petri dishes or other open media containers in the BACTROX for several weeks may lead to excessive drying of sample media. A small open container such as, a flask of 500ml of distilled water set on each shelf of the incubator can help to slow sample drying.

DATA OUTPUTS AND LOGGING

Each BACTROX hypoxia workstation is provided with a RS232 DB9 com port, an RS232 cable, and an RS232 to USB Type A adaptor. While the com port and cable are primarily intended for use in calibrating the oxygen sensor, the com port may also be used for data logging. Once per minute the BACTROX will send three outputs through the com port describing the current **temperature** (°C), **oxygen** (O₂), and **carbon dioxide** (CO₂) levels in the workspace chamber.

Use of the cable and com port requires downloading and installing the free, open source (**BSD license**) Tera Term terminal emulator on a laptop or other device. Tera Term can be downloaded from the **Tera Term development** project site (SourceForge.JP) at:

<http://en.sourceforge.jp/projects/ttssh2/releases/>

Using the RS232 to USB adaptor.

RS232 uses a communications protocol different from that of USB, and the adapter requires its own driver to convert between the two protocols. These drivers are available for free download on the adapter manufacture's website. Chose one compatible with your computer's operating system.

<http://www.iogear.com/support/dm/driver/GUC232A>

The BACTROX will not be able to communicate with a computer via the USB adapter without a driver.

See pages 57 – 58 in the O₂ Calibration procedure for instructions on how to connect the BACTROX to a logging computer, and how to initiate the logging process.

Logging Channels:

C1 = Current
Temperature

C3 = Current
Oxygen

C4 = Current
CO₂

OPERATION (CONTINUED)

MUTE THE AUDIBLE O₂ / CO₂ ALARMS

Visual O₂ or CO₂ deviation alarm indicators illuminate if gas levels deviate 1% above or below the current set points. An audible alarm for each gas type will sound immediately for high deviations. The low deviation audible alarms will sound only after the low indicator has been continually illuminated for 15 minutes. This delay prevents the alarm from sounding in the event a chamber entry creates a short-lived drop in gas concentration.



Figure 23:
Muted
Alarms

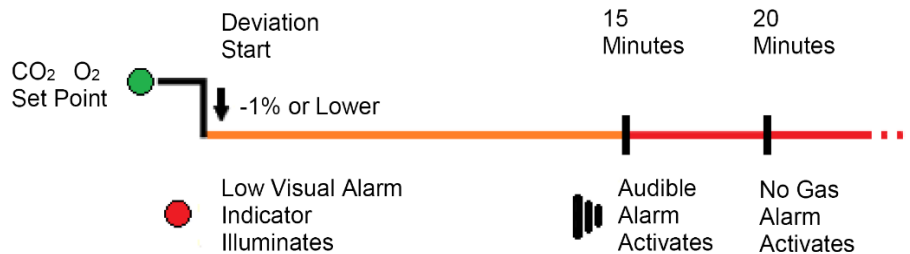
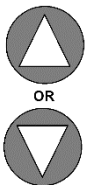


Figure 24: Gas Alarms Timeline



1. To mute an alarm, press and hold the **Up** or the **Down** arrow button on the control with the illuminated alarm indicator for 1 second.
2. The alarm will stay muted for the duration of the **current** temperature deviation, and the yellow Muted indicator will remain illuminated.
3. Another deviation of 1% will reactivate the audible alarm.

OPERATION (CONTINUED)

PRESSURE UNIT CONVERSION

Conversion table for pressure units

	kPa	MPa	kgf/cm ²	bar	psi	mmHg (Torr)	inHg	atm
1 kPa	1	1×10^{-3}	1.01972×10^{-2}	1×10^{-2}	1.45038×10^{-1}	7.50062	0.2953	9.86923×10^{-3}
1 MPa	1×10^3	1	1.01972×10	1×10	1.45038×10^2	7.50062×10^3	0.2953×10^3	9.86923
1 kgf/cm ²	9.80665×10	9.80665×10^{-2}	1	9.80665×10^{-1}	1.42234×10	7.35559×10^2	2.8959×10	9.67841×10^{-1}
1 bar	1×10^2	1×10^{-1}	1.01972	1	1.45038×10	7.50062×10^2	2.953×10	9.86923×10^{-1}
1 psi	6.89473	6.89473×10^{-3}	7.03065×10^{-2}	6.89473×10^{-2}	1	5.17147×10	2.036	6.80457×10^{-2}
1 mmHg (1 Torr)	1.33322×10^{-1}	1.33322×10^{-4}	1.35951×10^{-3}	1.33322×10^{-3}	1.93368×10^{-2}	1	3.9370×10^{-2}	1.31579×10^{-3}
1 inHg	3.3864	3.3864×10^{-3}	3.4531×10^{-2}	3.3864×10^{-2}	0.4912	2.5400×10	1	3.342×10^{-2}
1 atm	1.01325×10^2	1.01325×10^{-1}	1.03323	1.01325	1.46960×10	7.60000×10^2	2.9921×10	1

Figure 25: Pressure Measurement Unit Conversions

USER MAINTENANCE

Chamber Quality Control Check Sheet

Month:					
	Incubator Temperature	Condensate Drained	N ₂ Cylinder Pressure Reading	CO ₂ Cylinder Pressure Reading	
Date:					
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					
11					
12					
13					
14					
15					
16					
17					
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26					
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28					
29					
30					
31					

You may copy this sheet for institutional use

USER MAINTENANCE (CONTINUED)

DAILY MAINTENANCE

1. Record the gas cylinder reading(s) to track usage.
2. Change the gas cylinder(s) if low.
3. Remove and drain the condensate collection container as needed.
4. Visually inspect that the pass box gaskets are properly seated.
5. Check the cuffs on the sleeve system for holes, tears, and other signs of wear that may compromise integrity. Replace if necessary.
6. Check the incubator temperature to ensure that the setting is correct.
7. Change or reactivate the charcoal scrubber, if installed. Please see the [Activated Charcoal Scrubber and Rejuvenation Cycle](#) on page 44 in the Operation section.
8. Clean and disinfect the workspace chamber in accordance with your laboratory or production protocols, or regulatory requirements.

DOOR GASKET MAINTENANCE AND USAGE

BACTROX door gaskets are wear items. Replace when the gasket shows obvious signs of wear or damage such as, dryness, brittleness, cracks, or tears. Heavy institutional users may wish to keep a pair of spare door gaskets on hand (Part Number 3450507).

Spilling sample media on door gaskets or the interior surfaces of pass box doors may cause the gaskets to stick to the doors. This can compromise the atmospheric integrity of the pass box when door openings pull sticky gaskets of their mounts. Gaskets can be cleaned with dish soap and warm water, if compatible with your laboratory protocol.

SLEEVES MAINTENANCE AND USAGE

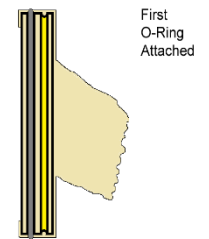
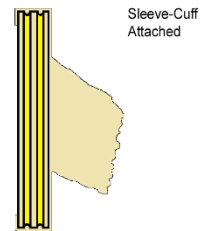
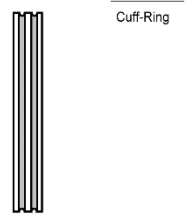
Sleeves may be washed with dish soap and warm water between uses. Disinfection should be carried out per lab protocols. Institutions with several users for each BACTROX may wish to keep a pair of sleeves in small, medium, and large sizes on hand. Or keep a pair of sleeves for each user.

The sleeve cuffs are subject to the most wear on the sleeve assemblies. Sheldon Manufacturing recommends keeping a stock of replacements on hand. See the [replacement parts list](#) (bottom of the list) on page 80 for sizes and part numbers of latex and nitrile cuffs. Make sure that the cuff matches the size of the sleeve assembly it will be attached to.

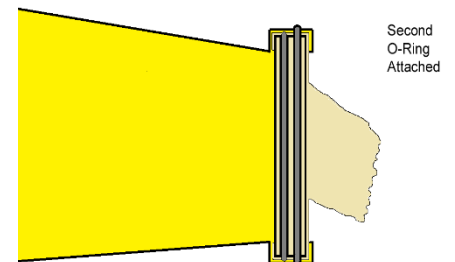
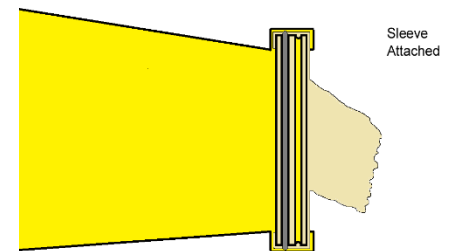
Sleeves or cuffs should be replaced when brittleness, dryness, or cracks are present.

REPLACING THE SLEEVE CUFFS

1. Remove the sleeve cuff by performing the following.
 - a. Remove the black O-ring located on the outside of the sleeve assembly, around the cuff. Gently roll the ring off to avoid damaging it or the sleeve.
 - b. Slowly and gently remove the sleeve-cuff and its sleeve cuff-ring from the sleeve.
 - c. Roll off the second black O-ring, which holds the sleeve-cuff to the cuff-ring.
 - d. Remove the old sleeve-cuff from the cuff-ring and discard. Inspect the cuff-ring. Replace the cuff-ring if there are cracks, dryness, brittleness, or a loss of flexibility.
 - e. Inspect the black O-rings. Replace if cracks, dryness, brittleness, or a loss of flexibility are present.
 - f. Inspect the sleeve. Replace if cracks, dryness, or brittleness are present.



2. Replace the sleeve-cuff.
 - a. Pull a new sleeve-cuff on over the cuff-ring.
 - b. Pull an O-ring onto the cuff-ring, over the sleeve-cuff. Fit the ring into the groove opposite the fringed side of the cuff.
 - c. Pull the lip of the sleeve over the cuff and cuff-ring. The fringed end of the cuff should be left outside the sleeve.
 - d. Roll on the second O-ring. Fit the O-ring into the unoccupied groove on the outside of the sleeve and sleeve cuff.



End of Procedure

Figure 25: Sleeve Cuff Replacement

USER MAINTENANCE (CONTINUED)



Warning: Prior to any maintenance or cleaning of this unit, disconnect the power cord from the power supply.

Avertissement: Avant d'effectuer toute maintenance ou entretien de cet appareil, débrancher le cordon secteur de la source d'alimentation.

CLEANING AND DISINFECTING

If a hazardous material or substance has spilled in the incubator, immediately initiate your site's Hazardous Material Spill Containment protocol. Contact your local Site Safety Officer and follow instructions per the site policy and procedures.

The unit should be cleaned and disinfected prior to first use. Periodic cleaning and disinfection are required to prevent microbiological contamination.

Do not use spray on cleaners or disinfectants. These can leak through openings and coat electrical components. Do not use cleaners or disinfectants that contain solvents capable of harming paint coatings or stainless steel surfaces. **Do not use chlorine-based bleaches or abrasives; these will damage the chamber liner.**



Warning: Never clean the unit with alcohol or flammable cleaners.

Avertissement: Ne jamais nettoyer l'appareil à l'alcool ou avec des nettoyeurs inflammables.

Note: Never spray the oxygen sensor or chamber power outlets. Do not attempt to clean or disinfect the interior of the O₂ sensor box cage. The high operating temperature of the sensor precludes microbiological contamination. Wipe down sensor and cage exterior surface with a lightly dampened cloth.

Cleaning

1. Remove all items (shelves, racks, and any additional items) from the incubator when possible.
2. Clean the workstation's interior with a mild soap and water solution, including all corners.
3. Clean all removable accessories including arm port doors, gaskets, and sleeve assemblies with a mild soap and water solution.
4. Rinse with distilled water and wipe dry with a soft cloth. **Do not use deionized water.** See the [Distilled and Deionized Water](#) paragraph on page 18 in the Operation section.
5. Take special care when cleaning around the oxygen sensor and chamber power outlets to prevent damage.
6. **Do not use chloride-based cleaners** except Zephiran benzalkonium chloride solution. Other types may have adverse effects on microbiological samples.
7. Wipe down the interior surfaces with Zephiran. Do not wipe up, allow the Zephiran to evaporate.

USER MAINTENANCE (CONTINUED)

Disinfecting

Disinfect the workspace chamber, pass box, and incubator on a regular basis. Perform the following steps to disinfect the workstation:

1. Turn the unit off. Carry out your laboratory disinfection protocol.
2. If possible, remove all interior accessories (shelf spacers, dish racks, and other non-attached items) from the incubator when disinfecting. Disinfect all corners, the incubator interior, and the pass box. Take special care when disinfecting around the pass box door and arm port door gaskets so as not to impair a positive seal.
3. Disinfect the unit using commercially available disinfectants that are non-corrosive, non-abrasive, and suitable for use on stainless steel surfaces. Contact your local Site Safety Officer for detailed information on the disinfectants compatible hypoxia applications.
4. **Do not use overtly volatile disinfecting agents.** Chlorines, amphyls, and quaternary ammonias will evaporate into the chamber environment. Over time the concentration in the chamber atmosphere will continue to increase, potentially leading to inhibited growth or metabolic symptoms in sample populations.
5. After completion of your institutional protocol, allow all disinfectants to evaporate completely. Wipe down all surfaces, except the acrylic glass panels, with distilled water and Zephiran until the unit no longer has a volatile odor. Use nonabrasive wipes.

MAINTAINING THE ACRYLIC GLASS PANELS

Sheldon Manufacturing recommends using Novus brand acrylic glass cleaner and Novus scratch remover for cleaning and maintaining BACTROX acrylic glass surfaces. Please see the [Accessories section](#) on page 80. Alcohol or alcohol-based solvents and other aggressive solvents should never be used to clean the BACTROX, and may damage the acrylic glass panels

Never expose the BACTROX to sustained UV light. Prolonged exposure to UV will result in rapid aging of the acrylic glass, leaving it vulnerable to compression forces, and generating cracks across all exposed areas. UV light will also quickly age sleeve assemblies, turning the sleeves yellow and result in a quick loss of elasticity.

Disable or redirect laboratory UV lighting away from the BACTROX. The BACTROX should not be exposed to direct sunlight. Verify that your laboratory or workspace environment does not use UV lighting at night. This type of light is usually referred to as short wave UVC or germicidal UV light, and operates at roughly 254nm.

Damage from prolonged or high intensity UV exposure is not covered under the manufacturing defects warranty.

Periodic use of long-wave (365nm) UV hand lamps used for bacterial identification should not damage the acrylic glass.

USER MAINTENANCE (CONTINUED)

CONDENSATION AND THE DEW POINT

Relative humidity inside the BACTROX should never exceed 80% at 25°C. Exceeding this threshold will likely result in condensation on incubator and workspace surfaces.

Condensation will appear wherever the humidity level in the chamber reaches the dew point. The dew point is the level of humidity at which the air cannot hold more water vapor. The warmer the air, the more water vapor it can hold.

As the level of humidity rises in the chamber, condensation will first appear on surfaces that are cooler than the air temperature. Near the dew point, condensation will form on any item or exposed surface that is even slightly cooler than the air. When the dew point is reached, condensation forms on nearly all exposed surfaces.

Mild condensation can be present in BACTROX units fully loaded or loaded to near capacity with open media plates, depending on ambient temperature and humidity. Cold air blowing on the exterior of the BACTROX may also help to cause condensation in the workspace chamber by chilling the acrylic glass panels and metal bulkheads.

Managing excessive condensation at humidity levels that overwhelm the BACTROX condensate controller depends on either lowering the humidity level in the chamber or increasing its air temperature.

Note: Note: Rising or falling air pressure from weather will adjust the dew point up and down in small increments. If the relative humidity in the BACTROX is already near the dew point, barometric fluctuations may push it across the dew point threshold.

If excessive condensation is forming in the BACTROX chamber, check the following:

- Check to see if condensate from the condensate controller is draining through the drain tube in the left wall and into the collection vessel.
- Remove or cap open containers of water or media. Empty the condensate controller catch vessel frequently.
- Does the number of media containers in the BACTROX exceed its rating? The workstation can hold 300 plates. Reduce the number of sample containers.
- Does the ambient humidity in the room exceed the BACTROX's stated operating range of 80% relative humidity? If so, lower the room's humidity.
- Is the BACTROX exposed to an external flow of cold air such as, an air-conditioning vent or a door to a cooler hallway or adjacent room? Block or divert the air, or move the incubator.
- Verify that the circulation fan over the Peltier condensate controller on left wall of the chamber is operating. The fan circulates air over condenser controller's cold plate.

ELECTRICAL COMPONENTS

Electrical components do not require maintenance. If the BACTROX electrical systems fail to operate as specified, please contact your distributor or Technical Support for assistance.

USER MAINTENANCE (CONTINUED)

Note: The O₂ sensor requires five minutes to warm up to its operating temperature in order to provide an accurate O₂ measurement. It will begin heating as soon as the BACTROX is turned on.

CALIBRATE THE O₂ SENSOR AND DISPLAY

The oxygen calibration procedures verify the accuracy of the O₂ display reading, and correct for any measurement errors. Gradual errors accrue from material changes in the sensor over its lifetime because of a high internal operating temperature, as well as exposure to ionized oxygen and other gasses. Additionally, the sensor is sensitive to altitude and changing barometric conditions when deriving O₂ concentration from the measured partial pressure of oxygen (PPO). Each BACTROX is shipped from the factory calibrated at near sea level for a two point curve using a N₂ – O₂ reference mixture of 5% O₂ and a near sea level ambient air sample near 20.7%.

Recommended calibrations

- Sheldon Manufacturing strongly recommends performing an oxygen sensor calibration procedure when preparing the unit for use at altitudes higher than 2000 feet (meters).
- During normal operations calibrate the O₂ sensor as often as required by your laboratory, study, or production protocol, or regulatory compliance requirements.

Chamber O₂ Deviations During Calibrations

The O₂ controller set point is set to off during calibration procedures. This means no N₂ or air injections take place during these procedures. The oxygen concentration in the chamber may drift from the set point during a calibration. Make sure to follow the entry and exit procedures when using the BACTROX arm ports during a calibration.

USER MAINTENANCE (CONTINUED)

Sample Types

Two air sample types may be used for calibrations.

- A reference mixed gas cylinder with a known O₂ concentration
- An ambient air sample with a calculated O₂ concentration

Reference gasses are typically a dry gas (no humidity) mix of nitrogen and a precise oxygen concentration. Reference gas mixes may be used for all calibration types.

The BACTROX may also be calibrated to ambient air drawn from the laboratory environment. To use an ambient air sample, it is necessary to calculate the oxygen concentration value of the air using the current temperature, humidity, and barometric pressure. The temperature and humidity should be obtained using sensors inside the laboratory environment in the immediate vicinity of the BACTROX. The current barometric pressure may be obtained from a local meteorological service or online source **unless** the BACTROX is located in an overpressure or negative pressure environment. In an artificial pressure environment, the air pressure must be measured in the same room as the BACTROX.

Please see the [Calculating the O₂ Concentration procedure](#) on page 84 for the formulas used to calculate the current ambient O₂ concentration. The ambient sample should be obtained by attaching the **air pump** provided with the BACTROX O₂ calibration kit to the BACTROX calibration port.



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Calibration Types

The BACTROX can be calibrated in one of two modes:

- Single Point Calibration
- Dual Point Calibration

Single Point Calibration: The single point calibration procedure produces a calibration offset adjustment to match the sensor and its display to a single calibration sample.

If a difference is detected between the sensor reading and the gas sample's known or calculated O₂ value, an offset adjustment is entered into the BACTROX controller. **The single point calibration and should only be used in applications in which having an absolute linear curve throughout your range of chamber O₂ concentrations is not an issue.** This may mean that a single point O₂ calibration is only valid for a single O₂ chamber concentration for your study.

Required for the single point calibration procedure:

- An ambient air sample or an oxygen reference cylinder
- The O₂ calibration kit provided with the BACTROX

USER MAINTENANCE (CONTINUED)

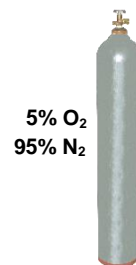
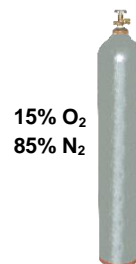
Note: The effective O₂ concentration operating range of the BACTROX is 1 – 20%

Note: The links and sites listed below are provided as conveniences. Sheldon Manufacturing makes no warrant as to the safety of third party sites, their contents, or the reliability or support of software products contained therein.

Dual Point Calibration: The dual point calibration procedure calibrates the sensor to an **upper calibration value point** and a **low calibration value point** to produce an adjusted linear curve across a range of potential chamber O₂ concentrations. The upper calibration may be made using either ambient laboratory atmosphere or a reference gas mix, and should never exceed 20.7%. **The low value is always calibrated using a hypoxic reference gas cylinder.**

Required for the dual point calibration procedure:

- An ambient air sample **or** an oxygen reference cylinder for the upper calibration point.
- A hypoxic oxygen reference mixture for the low calibration point.
- A computer connected to the BACTROX using the RS232 cable provided with the unit (includes USB adaptor).
- A software driver for the USB adaptor compatible with your computer operating system. RS232 uses a communications protocol different from that of USB, and the adapter requires its own driver to convert between the two protocols. These drivers are available for free download on the adapter manufacture's website:
 - <http://www.iogear.com/support/dm/driver/GUC232A>
- Terminal emulation software, such as, Tera Term, loaded on the computer (see below).
- The O₂ calibration kit provided with the BACTROX.



Dual Point O₂ Calibration Procedure

Note: Make sure the calibration kit cover plate is inside the workspace chamber prior to starting this procedure.

Prior to starting this procedure install a terminal emulator on the computer you will be using for the calibration and data logging. There are several free, open source emulators available online. Sheldon Manufacturing uses Tera Term when performing dual point calibrations and data logging at the factory. Tera Term is a free (**BSD license**) software application that can be downloaded from the **Tera Term development** project site (SourceForge.JP) at: <http://en.sourceforge.jp/projects/ttssh2/releases/>

Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Calibrating the O₂ Sensor

1. If you will be using an ambient air sample for the upper value calibration, calculate the current O₂ concentration of the ambient laboratory atmosphere.

- a. $WVP = \left(\frac{H_{rel}}{100}\right) \cdot WVP_{max}$

- b. $PPO_2 = (BP - WVP) \cdot \left(\frac{20.95}{100}\right)$

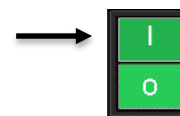
- c. $O_2\% = \left(\frac{PPO_2}{BP}\right) 100$

Please see the [Calculating the O₂ Concentration procedure](#) on page 84

2. Verify that the BACTROX has been powered for more than five minutes.
 - a. Calibrating with an O₂ sensor that has not warmed up fully will lead to an inaccurate calibration.



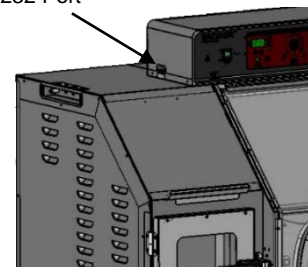
> 5 Minutes



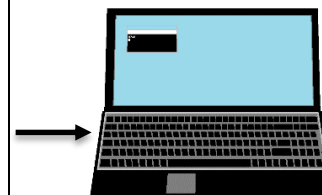
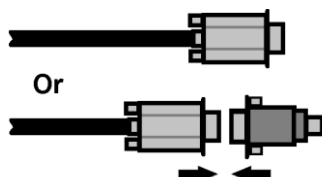
3. Connect the RS232 DB9 cable to the port on the left side of the BACTROX control panel box.



RS232 Port



4. Connect the RS232 DB9 cable to your computer.
 - a. Use the USB adaptor included with the calibration kit if necessary. Remember to download and install the free driver if using the adaptor.



USER MAINTENANCE (CONTINUED)

Calibrating the O₂ Sensor (Continued)

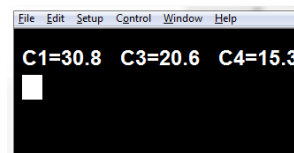
5. Open the terminal emulator on your computer.
 - a. For Tera Term, select the serial button in the lower left corner of the **New Connection** window.
 - b. If you know the communication port used by your computer, select it in the Port dropdown menu on the lower right side of the New Connection window.
 - c. If you do not know the com port, **select COM1**. The correct port may be determined by process of elimination. See next step (6).
 - d. **Click OK**.



6. Connection: If using Tera Term, the emulator should start logging data from the BACTROX once every minute if the correct com port was chosen. If the emulator is not logging the data, perform the following steps.



- a. Verify that a USB to Serial Adapter driver has been installed if you are using the RS232 to USB Adaptor
- b. Close Tera Term.
- c. Re-open Tera Term.
- d. Select a different com port.



Logging Channels:

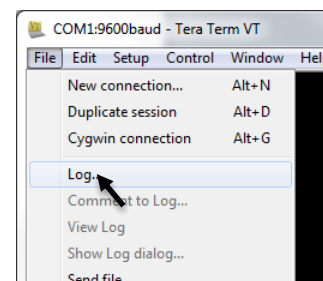
C1 = Current Temperature
C3 = Current Oxygen
C4 = Current CO₂

Note: The output logging channels and the calibration input channels are completely separate channels, with a separate numbering system.

7. Saving O₂ log data (optional). To save log data from the BACTROX during the calibration, perform the following steps.



- a. Click on the Tera Term File tab.
- b. Click on the "Log" option.
- c. Create a file on your computer to save the log data in.
- d. Click OK after naming the file.
- e. Data will now be saved.



Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Calibrating the O₂ Sensor (Continued)



OR



Wait 5 Seconds

8. Set the O₂ set point to OFF. This prevents the BACTROX controller from attempting to adjust the chamber O₂ concentration while the O₂ sensor is isolated from the workspace environment. It also disables the O₂ deviation alarms during the calibration process.

- Press and release either **Set Oxygen % arrow button**. The display will briefly flash the letters SP
- When the display dims and flashes an adjustable set point, press and hold the down arrow button until the display reads "Off".
- Wait five (5) seconds for the display to stop flashing.

SET OXYGEN %

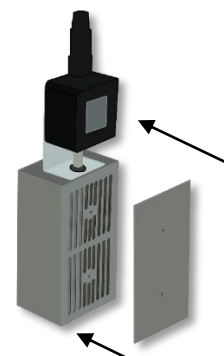


SET OXYGEN %



9. Install the O₂ calibration kit sensor cover plate on the oxygen sensor cage.

- Tighten the plate thumb screws until finger tight.
- This isolates the O₂ sensor and allows a sufficient concentration of calibration sample gas to build up around the sensor head.



Upper Value Calibration

10. Attach the upper calibration gas sample to the O₂ CAL PORT.



- If you are using **ambient air** for the upper calibration sample, plug the oxygen calibration pump included with the BACTROX into a suitable wall power source. Attach the calibration kit tubing to the pump and then to the O₂ CAL PORT on the BACTROX. Push the pump power switch to the (I) on position. The pilot light on the pump will illuminate, and air will be pumped into the O₂ sensor box.

Or

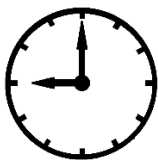

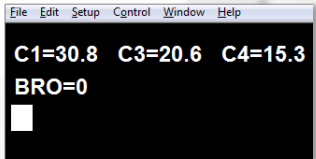




- If you are using a **reference gas mixture** for the upper calibration sample, connect the gas cylinder regulator to the O₂ CAL Port on BACTROX. Use the tubing provided with the oxygen calibration kit. Set the cylinder regulator to 3 PSI. **Never set the regulator to higher than 3 PSI!** Open the regulator control valve to establish a flow of gas to the sensor.



Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Calibrating the O ₂ Sensor (Continued)	
<p>11. Wait 5 minutes for a sufficient upper calibration O₂ sample concentration to build up in the sealed sensor box.</p>	 <p>Wait 5 Minutes</p>
<p>12. Access the BACTROX calibration channels using the terminal emulator software on your computer after 5 minutes have elapsed.</p> <ol style="list-style-type: none"> Type BRO=0 in the emulator window and press Enter. You now have access to the calibration channels.  <p>Note: The BACTROX will automatically revert out of the calibration channels every three minutes to prevent accidental calibration adjustments from being made.</p>	
<p>Note: The 20.7% shown to the right is an arbitrary example value.</p> <p>13. Check the green SET OXYGEN % display on the BACTROX.</p> <ol style="list-style-type: none"> If BACTROX O₂ display matches the O₂ concentration of your upper calibration sample, the upper value is now calibrated. Advance to step 16. <p style="text-align: center;">Or</p> <p>14. If there is a difference between the O₂ display reading and the concentration of your sample, and that difference exceeds the range allowed by your laboratory protocol, enter an upper calibration adjustment. See next step.</p>	<p>Upper Sample O₂% = 20.7%</p> <p>SET OXYGEN %</p>  <p style="text-align: center;">Or</p> <p>SET OXYGEN %</p> 

Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Calibrating the O₂ Sensor (Continued)

15. Enter an upper calibration adjustment to match the Set Oxygen % display to the upper calibration gas sample value.

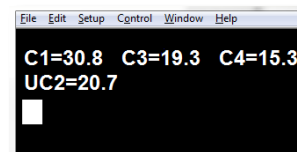


- Enter the air or reference gas concentration into the BACTROX by typing **UC2=sample value** in the emulator window. For example, if your air sample or reference gas has an O₂ concentration of 20.7%, type **UC2=20.7** and press **Enter**.
- The BACTROX display should now match the O₂ concentration of your sample.

Note: UC2 indicates the upper calibration value for oxygen

Upper Sample O₂% =
20.7%

SET OXYGEN %



SET OXYGEN %





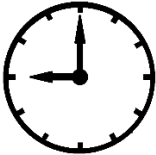


16. Turn off and unplug the calibration kit air pump, or close the valve of the reference gas cylinder. Set the cylinder regulator to 0. Disconnect the pump or cylinder from the O₂ Cal Port.



Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Low Value Calibration

Calibrating the O ₂ Sensor (Continued)	
 <p>17. Attach your low value reference gas mixture to the O₂ Cal Port. Do not use ambient air for the low value sample.</p> <ol style="list-style-type: none">Set the cylinder regulator to 3 PSI. Do not set the regulator to higher than 3 PSI!Open the regulator control valve to establish a flow of gas to the sensor.	
<p>18. Wait 5 five minutes for the low value reference mix to purge the sensor box and establish a sufficient concentration.</p>	 <p>Wait 5 Minutes</p>
<p>Note: The 5% shown to the right is an arbitrary example value. The O₂ value of your low value gas mix should be chosen on the basis of your study or laboratory protocol requirements.</p> <p>19. Check the SET OXYGEN % display on the BACTROX.</p> <ol style="list-style-type: none">If the display matches the O₂ concentration of the low calibration gas mix, the BACTROX low calibration value is now calibrated. Advance to step 21. <p style="text-align: center;">Or</p> <ol style="list-style-type: none">If there is a difference between the display and the reference gas concentration, and that difference exceeds the acceptable range of your laboratory or study protocol, enter a low calibration adjustment. See next step.	<p>Lower Sample O₂% = 5.0%</p> <p>SET OXYGEN %</p>  <p>Or</p> <p>SET OXYGEN %</p> 

Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Calibrating the O₂ Sensor (Continued)

20. Enter a low calibration adjustment to match the O₂ sensor and display to the sample concentration value.

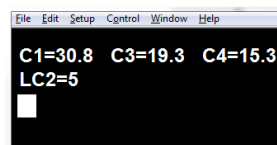


- Type **BRO=0** in the Tera Term window and press **Enter** to ensure that you are still accessing the calibration channels.
- Enter the low calibration gas O₂ concentration by typing **LC2=sample value** in the Tera Term window. For example, if the low calibration reference gas has an O₂ concentration of 5%, **type LC2=5** and hit **Esnter**.
- The BACTROX display should now match the O₂ concentration value of the low calibration reference gas.

Note: LC2 indicates the low calibration value for oxygen

Lower Sample O₂%
= 5.0%

SET OXYGEN %



SET OXYGEN %



Verify Calibration Across Full Range

21. Verify that the sensor is accurate across the full range of the established O₂ curve by rechecking the high calibration accuracy.

- Close the valve and set the regulator of the low reference gas cylinder to zero (0). Disconnect the low cylinder from the calibration gas port.
- Re-attach your upper calibration sample to the O₂ Cal Port. Plug in and turn on the calibration kit air pump. **Or**, set the upper reference cylinder regulator to 3 PSI, and open the regulator control valve to establish a flow of gas to the sensor.
- Wait five 5 minutes for the upper calibration sample gas to purge the sensor box and build up a sufficient concentration. Failure to wait for the full 5 minutes will result in an inaccurate calibration.



Wait 5 Minutes

Low Value





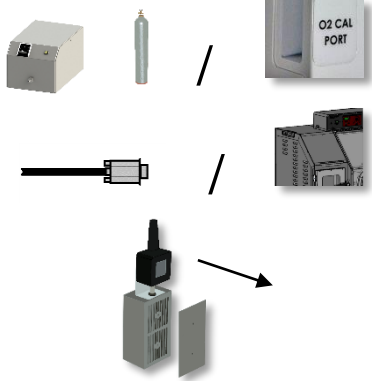

Upper Value



Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Verification of Range Continued

Calibrating the O ₂ Sensor (Continued)	
<p>22. If the BACTROX display matches the O₂ concentration of the upper calibration sample, the dual point calibration is accurate across the range of the curve. Advance to step 23.</p> <ol style="list-style-type: none"> If the O₂ concentration does not match the concentration of the air or reference gas sample, repeat steps 10 – 21 starting on page 60. Recalculate the ambient air O₂ concentration if you are using ambient air for the high calibration. Contact Sheldon Manufacturing Technical Support for assistance if the high value does not match the ambient air or reference gas concentration after three attempts. 	<p>Upper Sample O₂% = 20.7%</p> <p>SET OXYGEN %</p>  <p>Or</p> <p>SET OXYGEN %</p>  <p>Repeat steps 10 – 21</p>
<p>23. Return the unit to normal operation after a successful calibration.</p> <ol style="list-style-type: none"> Disconnect the calibration air pump or high calibration gas cylinder. Reattach the O₂ Cal Port cover cap. Disconnect the RS232 cable unless you wish to continue logging data from the BACTROX. Remove the calibration kit cover plate from the oxygen sensor! Failure to do so will result in inaccurate O₂ readings. Set the O₂ concentration set point (page 37) for your application using the Set Oxygen % arrow keys. The oxygen concentration in the workspace chamber may have fallen or risen during the calibration procedure. Monitor the BACTROX and make sure it achieves the set point. Gas injection lights will illuminate as the unit matches and stabilizes at the set point. . 	 <p>SET OXYGEN %</p>  <p>↓</p> <p>Your Application O₂% Set Point</p>


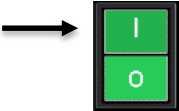





End of dual point calibration

USER MAINTENANCE (CONTINUED)

Single Point O₂ Calibration

A single point calibration is valid for a narrow range centered on the calibration sample value. For example, if the unit is calibrated using a reference gas with 10% O₂, the calibration is valid for a narrow band around 10%. Sheldon Manufacturing recommends using a calibration reference gas with an O₂ concentration that matches your application set point.

Note: Make sure the calibration kit cover plate is inside the workspace chamber prior to starting this process.

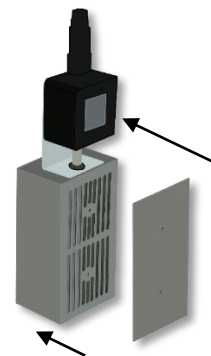
Calibrating the O ₂ Sensor	
<p>1. Verify that the BACTROX has been powered for more than five minutes.</p> <ul style="list-style-type: none">a. Calibrating with an O₂ sensor that has not warmed up fully will lead to an inaccurate calibration.	 <p>> 5 Minutes</p> 
<p> OR </p> <p>2. Set the O₂ set point to OFF. This prevents the BACTROX controller from attempting to adjust the chamber O₂ concentration while the O₂ sensor is isolated from the workspace environment. It also disables the O₂ deviation alarms during the calibration process.</p> <ul style="list-style-type: none">a. Press and release either Set Oxygen % arrow button. The display will briefly flash the letters SPb. When the green Oxygen % display dims and flashes an adjustable set point, press and hold the down arrow button until the display reads "Off".c. Wait five (5) seconds for the display to stop flashing.  <p>Wait 5 Seconds</p>	<p>SET OXYGEN %</p>  <p>↓</p> <p>SET OXYGEN %</p> 

Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Single Point O₂ Sensor (Continued)

3. Install the O₂ calibration kit sensor cover plate on the oxygen sensor cage.
 - a. Tighten the plate thumb screws until finger tight.
 - b. This isolates the O₂ sensor and allows a sufficient concentration of calibration sample gas to build up around the sensor head.



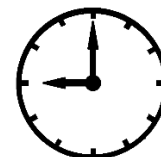
4. Attach the calibration gas sample to the O₂ CAL PORT.



- a. Connect the gas cylinder to the O₂ CAL Port on BACTROX.
- b. Use the tubing provided with the oxygen calibration kit.
- c. Set the cylinder regulator to 3 PSI. **Never set the regulator to higher than 3 PSI!** Open the regulator control valve to establish a flow of gas to the sensor.



5. Wait five (5) minutes for a sufficient sample concentration to build up in the sealed sensor box.



Wait 5 Minutes

Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Single Point O₂ Sensor (Continued)

Note: The 15% O₂ sample value and 13.4% display are arbitrary example values.

6. Check the green SET OXYGEN % display.
 - a. If display matches the O₂ concentration of your calibration reference mix, the BACTROX is now calibrated for O₂. Advance to step 9.
 - Or**
 - b. If there is a difference between the display reading and the concentration of your sample, and that difference exceeds the range allowed by your laboratory protocol, enter a calibration offset. **See next step.**

Sample O₂% = 15%

SET OXYGEN %



Or

SET OXYGEN %



7. Place the display in its calibration mode.



AND



- a. Press and hold both the **UP and DOWN** Set O₂ arrow buttons simultaneously for approximately 5 seconds.
- b. Release the buttons when the display shows the letters "CO". The display will begin flashing the **current O₂ display value**.

Sample O₂% = 15%

SET OXYGEN %



Note: If an arrow key is not pressed for five seconds, the display will cease flashing, and store the last displayed value as the new current O₂ value.



AND



8. Use the **Up** or **Down** arrows to adjust the current O₂ display value until it matches the reference gas O₂ concentration.

SET OXYGEN %



9. After matching the display to the reference device, wait 5 seconds.







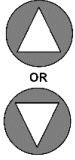
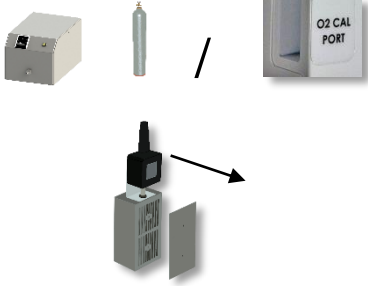


Wait 5 Seconds

- a. The display will cease flashing and store the corrected display value.
- b. The workstation will begin injecting O₂ or allow the current gas concentration to decay in order to achieve the set point with the corrected display value.

SET OXYGEN %



USER MAINTENANCE (CONTINUED)

Single Point O ₂ Sensor (Continued)	
<p>10. Allow the BACTROX to sit for at least 1 hour undisturbed to stabilize after it has achieved the corrected O₂ set point.</p>  <p>Wait 1 Hour</p> <p>a. Failure to wait until the unit is fully stabilized will result in an inaccurate reading and calibration.</p>	<p>SET OXYGEN %</p> 
<p>11. If the BACTROX display matches the O₂ concentration of the reference gas, the O₂ sensor and display are now calibrated. Advance to step 10.</p> <p style="text-align: center;">Or</p> <p>a. If the O₂ concentration does not match the concentration of the reference gas sample, repeat steps 6 - 9 starting on page 66.</p> <p>b. Contact Sheldon Manufacturing Technical Support for assistance if the display value does not match your sample concentration after three attempts.</p>	<p>Sample O₂% = 15%</p> <p>SET OXYGEN %</p>  <p style="text-align: center;">Or</p> <p>SET OXYGEN %</p>  <p style="text-align: center;">Repeat steps 6 - 11</p>
<p>12. Return the unit to normal operation after a successful calibration.</p> <p>a. Disconnect the calibration air pump or reference gas cylinder.</p> <p>b. Reattach the O₂ Cal Port cover cap.</p> <p>c. Remove the calibration kit cover plate from the oxygen sensor.</p> <p>d. Set the O₂ concentration set point (see page 37) for your application using the Set Oxygen % arrow keys.</p> <p>e. The oxygen concentration in the workspace chamber may have fallen or risen during the calibration procedure. Monitor the BACTROX and make sure it returns to the set point. Gas injection lights will illuminate as the unit matches and stabilizes at the set point.</p> 	 <p>SET OXYGEN %</p>  <p style="text-align: center;">↓</p>  <p style="text-align: center;">Your Application O₂% Set Point</p>

End of procedure

USER MAINTENANCE (CONTINUED)

CALIBRATE THE CO₂ DISPLAY

Note: Performing a CO₂ display calibration requires a gas reference device. Please see the [Reference Sensor Devices entry](#) on page 11 for the device requirements.

CO₂ calibrations are performed to match the incubator CO₂ display to the actual gas concentration in the workspace chamber. The actual concentration is supplied by a calibrated reference sensor device. Calibrations compensate for drifts in the unit microprocessor controller, as well as those caused by the natural material evolution of the IR CO₂ sensor when continually exposed the chamber atmosphere. Calibrate as often as required by your laboratory or production protocol, or regulatory compliance schedule.

CO₂ Supply

The incubator must be powered, the CO₂ set point set, and the BACTROX supplied with CO₂ for at least eight hours prior to the calibration

Temperature

The chamber temperature impacts the chamber CO₂ concentration, due to its influence on gas diffusion. CO₂ calibrations must be performed with the incubator fully heated and stable at your operational temperature set point. A CO₂ display calibration may be performed during a temperature calibration **as long as pass box or armor port doors are not opened during either procedure.**

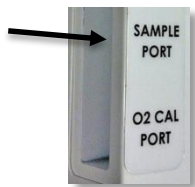


Figure 26:
CO₂ Sample
Port

Probes








Connect a digital CO₂ analyzer to the sample tube to the sample port, located on the left side of the incubator near the top.

Stability

Prior to a calibration the chamber must operate at its CO₂ set point for **at least 1 hour with no fluctuations** of $\pm 0.1\%$ or greater in order to be considered stabilized. Failure to wait for stabilization will result in an inaccurate calibration and incubator display reading.

For best results, allow the unit to operate undisturbed for 8 hours supplied to achieve temperature and gas stability (for example, overnight).

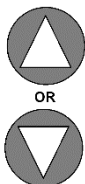
USER MAINTENANCE (CONTINUED)

Calibrate the CO ₂ Display	
<p>1. Once the BACTROX CO₂ concentration has stabilized with no fluctuations of 0.1% or greater, compare the gas reference device and BACTROX CO₂ display readings.</p> <ol style="list-style-type: none"> If the readings are the same, or the difference between the two (2) falls within the acceptable range of your protocol, the display is accurately showing the chamber CO₂ concentration. The CO₂ calibration procedure is now complete. If there is a difference between the two readings that falls outside the acceptable range of your protocol see the next step. 	<p>Reference Device</p>  <p>Set CO₂</p> 
<p>2. A display calibration adjustment must be entered to match the incubator CO₂ display to the reference device.</p>	<p>Reference Device</p>  <p>Set CO₂</p> 
<p>3. Place the display in its CO₂ calibration mode.</p> <div style="display: flex; align-items: center;">  <div style="margin-left: 10px;"> <p>a. Press and hold both the UP and DOWN Set CO₂ arrow buttons simultaneously for approximately 5 seconds.</p> </div> </div> <div style="display: flex; align-items: center;">  <div style="margin-left: 10px;"> <p>b. Release the buttons when the display shows the letters "CP". The display will begin flashing the current CO₂ display value.</p> </div> </div> <p>Note: If an arrow key is not pressed for five seconds, the display will cease flashing, and store the last displayed value as the new current chamber CO₂ value.</p>	<p>Set CO₂</p> 

Procedure continued on next page

USER MAINTENANCE (CONTINUED)

Calibrate the CO₂ Display (Continued)



4. Use the **Up** or **Down** arrows to adjust the current CO₂ display value until it matches the reference device CO₂ reading.

Reference Device

8 5.8 %

Set CO₂

8 5.8 ✓

5. After matching the display to the reference device, wait 5 seconds.



Wait 5 Seconds

a. The display will cease flashing and store the corrected display value.

b. The workstation will begin injecting CO₂ or allow the current gas concentration to decay in order to achieve the set point with the corrected display value.

Set CO₂

8 5.6



Wait 1 Hour

6. Allow the BACTROX to sit for at last 1 hour undisturbed to stabilize **after it has achieved the corrected CO₂ set point**.

a. Failure to wait until the unit is fully stabilized will result in an inaccurate reading and calibration.

Set CO₂

8 5.0

7. Compare the reference device reading with the BACTROX CO₂ display again.

a. If the reference device and the CO₂ display readings are the same or the difference now falls within the range of your protocol, **the incubator is now calibrated for CO₂**.

b. Or see next step.

Reference Device



8 5.0 %

Set CO₂

8 5.0 ✓

Procedure continued on next page

USER MAINTENANCE (CONTINUED)

CO ₂ Calibration (Continued)	
<p>8. If the two readings are not the same, and the difference falls outside the acceptable range of your protocol, repeat steps 3 – 7 up to two more times.</p> <p>a. Three calibration attempts may be required to successfully calibrate units that are more than $\pm 2\%$ out of calibration.</p>	<p>Reference Device</p>  <p>Set CO₂</p> 
<p>9. If the CO₂ readings of the display and the reference device still fall outside your protocol after three calibration attempts, contact your distributor or Sheldon Technical Support for assistance.</p> <p>a. Three calibration attempts may be required to successfully calibrate units that are more than $\pm 2\%$ out of calibration.</p>	

End of procedure

USER MAINTENANCE (CONTINUED)

CALIBRATE THE TEMPERATURE DISPLAY

Note: Performing a temperature display calibration requires a temperature reference device. Please see the [Reference Sensor Devices entry](#) on page 11 for device requirements.

Temperature calibrations are performed to match the temperature display to the actual air temperature inside the incubation chamber. The actual air temperature is supplied by a calibrated reference sensor device. Calibrations compensate for drifts in the unit microprocessor controller as well as those caused by the natural material evolution of the sensor probe in the humidified and heated chamber space. Calibrate as often as required by your laboratory or production protocol, or regulatory compliance schedule.

Humidity

Always calibrate for temperature with the chamber humidified.

Probes

Reference device sensing probes may be run through either finger hole in the incubator doors. Probes may also be introduced through the chamber door space. Use non-stick, non-marking tape to secure the wires and probe heads, and seal any gaps in the doorspace created by a probe wire.

Place the sensor probe of the temperature reference device inside as close as possible to the geometric center of the chamber. A thermocouple sensor probe sleeve may be taped to the shelving, as long as the exposed copper end is 2 inches (5cm) away from the shelf (see Figure 25). An exposed sensor probe in direct contact with the shelving may experience heat sinking, which can result in an inaccurate temperature reading.

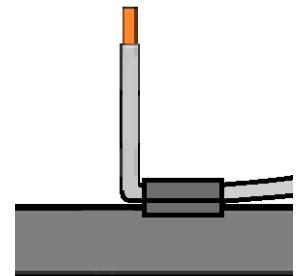


Figure 27: Probe End 2 inches (5cm) From Shelf Surface

Stability

Prior to a calibration the incubator should operate undisturbed at its application temperature set point for 8 hours in order to stabilize. A common practice is to introduce and place the temperature sensor probe in the incubator, allow the unit to operate and stabilize overnight, and then conduct the calibration the next day.



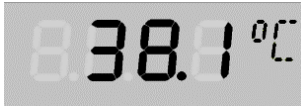






Wait 1 Hour

The incubator is considered stabilized when it has operated for **1 hour** with no fluctuations $\pm 0.1^{\circ}\text{C}$ or greater. Failure to wait for stabilization will result in an inaccurate calibration.











Continued on next page

USER MAINTENANCE (CONTINUED)

Temperature Calibration	
<p>1. Once the incubator temperature has stabilized, compare the reference device and BCTROX temperature display readings.</p> <p>a. If the readings are the same, or the difference between the two (2) falls within the acceptable range of your protocol, the display is accurately showing the test space chamber air temperature. The Temperature Calibration procedure is now complete.</p> <p>b. If a difference fall outside of your protocol range, advance to step 2.</p>	<p>Reference Device</p>  <p>Set Temperature °C</p> 
<p>2. A display calibration adjustment must be entered to match the display to the reference device. See next step.</p>	<p>Reference Device</p>  <p>Set Temperature °C</p> 
<p>3. Place the display in its temperature calibration mode.</p> <p> AND </p> <p>a. Press and hold both the UP and DOWN temperature arrow buttons simultaneously for approximately 5 seconds.</p> <p>b. Release the buttons when the temperature display shows the letters "CP". The display will begin flashing the current temperature display value.</p> <hr/> <p>Note: If an arrow key is not pressed for five seconds, the display will cease flashing, and store the last displayed value as the new current chamber temperature value.</p>	<p>Set Temperature °C</p> 



Continued on next page

USER MAINTENANCE (CONTINUED)

Temperature Calibration (Continued)	
 <p>OR</p>  <p>4. Use the Up or Down arrows to adjust the current display temperature value until it matches the reference device temperature reading.</p>	<p>Reference Device</p>  <p>Set Temperature °C</p> 
<p>5. After correcting for the difference, wait five (5) seconds.</p>  <p>Wait 5 Seconds</p> <p>a. The temperature display will cease flashing and store the corrected chamber display value.</p> <p>b. The incubator will now begin heating or passively cooling in order to reach the set point with the corrected display value.</p>	<p>Set Temperature °C</p>  <p>Adjusting to Set Point</p>
 <p>Wait 1 Hour</p> <p>6. Allow the BACTROX sit for at last one 1 hour undisturbed to stabilize after the incubator has achieved the corrected temperature set point.</p> <p>a. Failure to wait until the incubator is fully stabilized will result in an inaccurate reading.</p>	<p>Set Temperature °C</p> 
<p>7. Compare the reference device reading with the chamber temperature display again.</p> <p>a. If the reference device and the chamber temperature display readings are the same or the difference falls within the range of your protocol, the incubator is now calibrated for temperature.</p> <p>b. See the next step if the readings fail to match or fall outside of your protocol range.</p>	<p>Reference Device</p>  <p>Set Temperature °C</p> 

Continued on next page

USER MAINTENANCE (CONTINUED)

Temperature Calibration (Continued)	
<p>8. If the two readings are not the same, and the difference still falls outside the acceptable range of your protocol, repeat steps 3 – 7 up to two more times.</p> <p>a. Three calibration attempts may be required to successfully calibrate units that are more than $\pm 2^{\circ}\text{C}$ out of calibration.</p>	<p>Reference Device</p>  <p>Set Temperature $^{\circ}\text{C}$</p> 
<p>9. If the temperature readings of the chamber and the reference device still fall outside your protocol after three calibration attempts, contact your distributor or Sheldon Technical Support for assistance.</p>	

End of procedure

UNIT SPECIFICATIONS

The BACTROX is a 110 – 120 volt workstation. Please refer to the BACTROX data plate for individual electrical specifications. Please refer to the BACTROX data plate for individual electrical specifications.

Technical data specified applies to units with standard equipment at an ambient temperature of 25°C (77°F) and a voltage fluctuation of ±10%. The temperatures specified are determined in accordance to factory standard following DIN 12880 respecting the recommended wall clearances of 10% of the height, width, and depth of the inner chamber. All indications are average values, typical for units produced in the series. We reserve the right to alter technical specifications at all times.

WEIGHT

Shipping Weight	Unit Weight
480lbs / 218kg	247 / 112kg

WORKSTATION DIMENSIONS

By Inches

Exterior W x D x H	Workspace Chamber W x D x H
48.7 x 31.3 x 32.5 inches	33 x 28.5 x 25.2 inches

By Centimeters

Exterior W x D x H	Interior Chamber W x D x H
124 x 79.5 x 82.5 cm	83.8 x 72.4 x 64 cm

STAND DIMENSIONS

Optional Stand Available for Purchase

Inches W x D x H	Centimeters W x D x H
49 x 30 x 30 inches	124.5 x 76.2 x 76.2cm

UNIT SPECIFICATIONS (CONTINUED)

PASS BOX CAPACITY

Interior Dimensions

Inches W × D × H	Centimeters W × D × H
9 x 10.7 x 9 inches	23 x 27.5 x 23 cm

Pass Box Volume

Cubic Feet	Liters
0.5	14

Total Plate Capacity

Plates
300

WORKSPACE CAPACITY

Workspace Chamber Volume

Cubic Feet	Liters
13.7	388

Workspace Incubator Volume

Cubic Feet	Liters
1.7	48

Workspace Chamber Incubator Interior

Inches W × D × H	Centimeters W × D × H
27.5 x 8 x 13.5	70 x 20 x 34

UNIT SPECIFICATIONS (CONTINUED)

INCUBATOR TEMPERATURE

Range	Uniformity
Ambient +8°C to 60°C	±0.35°C @ 37°C

GAS RANGES

O ₂ Concentration	CO ₂ Concentration
0.5 – 20%	1 – 20%

POWER

Model	AC Voltage	Amperage	Frequency
BACTROX	110-120	3.5	50/60 Hz

PARTS LIST

Description	Parts Number
Pass Box Door Gasket 9 x 9 burgundy	3450507
Arm Port Door Left	9521253
Arm Port Door Right	9521254
Arm Port Door O-Ring	6000509
Fuse 4A 5 X 20 Type T Slow Blow	3300537
Leveling Foot	2700506
Petrie Dish Rack 2 x 11 inches (5 X 28cm)	5110729
Power Cord 5-15 NEMA 15A	1800510
Rubber Sleeve Cuff Plugs	7200502
Shelf Spacer	5680502
Sleeve Pair 10 inches (25 cm)	3600521
Sleeve Assembly Size 8 Medium (2 cuffs, 2 cuff-rings, 4 0-rings, 2 sleeves, 2 straps)	9990738M
Sleeve Assembly) Size 9 Large (2 cuffs, 2 cuff-rings, 4 0-rings, 2 sleeves, 2 straps)	9990738L
Sleeve Assembly) Size 6.5 Extra Small (2 cuffs, 2 cuff-rings, 4 0-rings, 2 sleeves, 2 straps)	9990738XS
Sleeve Cuff-Ring 4 Inches (interior diameter)	6400590
Sleeve Cuff-Ring Black O-ring	6000504
Sleeve Cuff-Ring 3.5 Inches (interior diameter) Small	6400619
Sleeve Cuff-Ring Red O-ring	6000503
Sleeve Cuffs Latex (for sleeve assembly) size 7	3600500
Sleeve Cuffs Latex (for sleeve assembly) size 8	3600501
Sleeve Cuffs Latex (for sleeve assembly) size 9	3600502
Sleeve Cuffs Nitrile (for sleeve assembly) size 7	3600513
Sleeve Cuffs Nitrile (for sleeve assembly) size 8	3600514
Sleeve Cuffs Nitrile (for sleeve assembly) size 9	3600515

PARTS LIST (CONTINUED)

ORDERING PARTS AND CONSUMABLES

If you have the Part Number for an item, you may order it directly from Sheldon Manufacturing by calling 1-800-322-4897 extension 3. If you are uncertain that you have the correct Part Number, or if you need that specific item, please contact Sheldon Technical Support for help at 1-800-322-4897 extension 4 or (503) 640-3000. Please have the **model number** and **serial number** of the BACTROX ready, as Tech Support will need this information to match your workstation with its correct part.

ACCESSORIES

Shel Lab offers the following BACTROX accessories for sale. Please see our website for prices.

Acrylic Glass Cleaner (8 oz)

Novus brand acrylic / plastic glass cleaner.

Part Number 1060503



Acrylic Glass Scratch Remover (2 oz)

Helps remove visible scratches and nicks from acrylic glass.

Part Number 1060504



Anatox Activated Charcoal (2 lbs / 0.9 kgs)

For scrubbing hydrogen sulfides, fatty acids, and some toxic or corrosive compounds from the workstation's atmosphere.

Part Number 1060500.



Anatox Fan

Holds Anatox charcoal filters and significantly speeds the removal of sulfides, fatty acids, and toxic or corrosive compounds.

Part Number 9490578



Compressed Gas Cylinder Switcher, Automatic

Allows two gas cylinders to be connected to one gas port, and automatically switches from the first to the second cylinder when the first is empty.

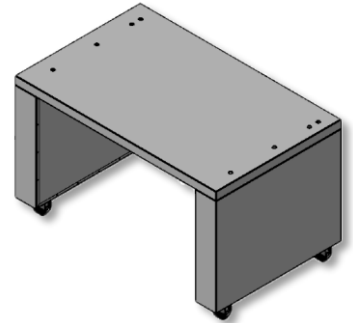
Part Number 2002-B



ACCESSORIES (CONTINUED)

BACTROX: BAC1 STAND

A rolling stand with cabinet for the BACTROX
30 inches high, 49 inches wide, 30 inches depth
(76.2cm high by 124.5 cm wide by 76.2cm depth)
Part Number BAC1 STAND (9000510)



Lukas Fiber Optic Micro Lite Illumination System

A fiber optic, adjustable brightness, halogen light box and guide. Provides a stable, long lasting light for use with BACTROX workstations and stereo microscopes.
Part Number 4650503



UV Viewing Lamp

A handheld UV lamp for use with BACTROX workstations.
Parts Number 9490507



Zephiran Benzalkonium Chloride Chamber Cleaner

1 Gallon, 0.133%.
Part Number 1060501



APPENDIX

Note: It is not necessary to calculate the O₂ concentration of the laboratory atmosphere if you will be using O₂ reference gas mixes for calibration. Calculating the ambient O₂% is only required when calibrating to an ambient atmosphere sample.

CALCULATING THE O₂ CONCENTRATION

To determine the oxygen concentration of the ambient laboratory atmosphere it is necessary to find the partial pressure of the oxygen (PPO) in the atmosphere. To calculate the PPO, it's first necessary to calculate the water vapor pressure (WVP) and then subtract it from the current barometric pressure.

Calculations

1. Water Vapor Pressure in millibars (mbar)
2. Partial Pressure O₂ in millibars (mbar)
3. O₂ Concentration (%)

1) Water Vapor Pressure (WVP) in mbars

$$WVP = \left(\frac{H_{rel}}{100} \right) \cdot WVP_{max}$$

Calculating the Water Vapor Pressure requires:

- **Relative Humidity (H_{rel})** of the laboratory ambient atmosphere
 - The relative humidity of the laboratory atmosphere can be obtained using a variety of commercially available handheld or desktop sensors. The sensor should provide a relative humidity value accurate to ±1%.
- **Temperature** of the laboratory atmosphere
 - Temperature is used to look up the Water Vapor Pressure Max (WVP_{max}). The thermometer device should measure the temperature of the airspace near the BACTROX. **Do not use** the room's climate control thermostat setting or display reading for this value. Both may vary significantly from the air temperature around the BACTROX.
- **Water Vapor Pressure Maximum (WVP_{max})**
 - Please see the WVP_{max} for table on the next page to find Water Vapor Pressure Max for your current air temperature.

Note: Never use a relative humidity value taken from outside the room the BACTROX is located in. Use of relative humidity from another room or a weather report will result in an inaccurate O₂ sensor calibration.

APPENDIX (CONTINUED)

Water Vapor Pressure Max (WVP_{max})¹

Temperature (C°)	WVP _{max} (mbar)
15	17.06
16	18.19
17	19.38
18	20.65
19	21.98
20	23.39
21	24.88
22	26.45
23	28.10
24	29.85
25	31.69
26	33.62
27	35.66
28	37.81
29	40.07
30	42.44

2) Calculating the PPO₂ in mbars

After calculating the WVP value, calculate the Partial Pressure Oxygen for the laboratory environment.

$$PPO_2 = (BP - WVP) \cdot \left(\frac{20.95}{100}\right)$$

Calculating the PPO₂ requires:

- **Water Vapor Pressure**
 - The WVP value calculated in step 1 on the previous page
- **Barometric Pressure (BP) in millibars (mbars)**
 - The current Barometric Pressure for your area can be obtained from a local metrological station or online weather service. If the BACTROX is located in an overpressure or negative pressure laboratory environment, it will be necessary to obtain a barometric pressure sensor to determine the BP value for the room.

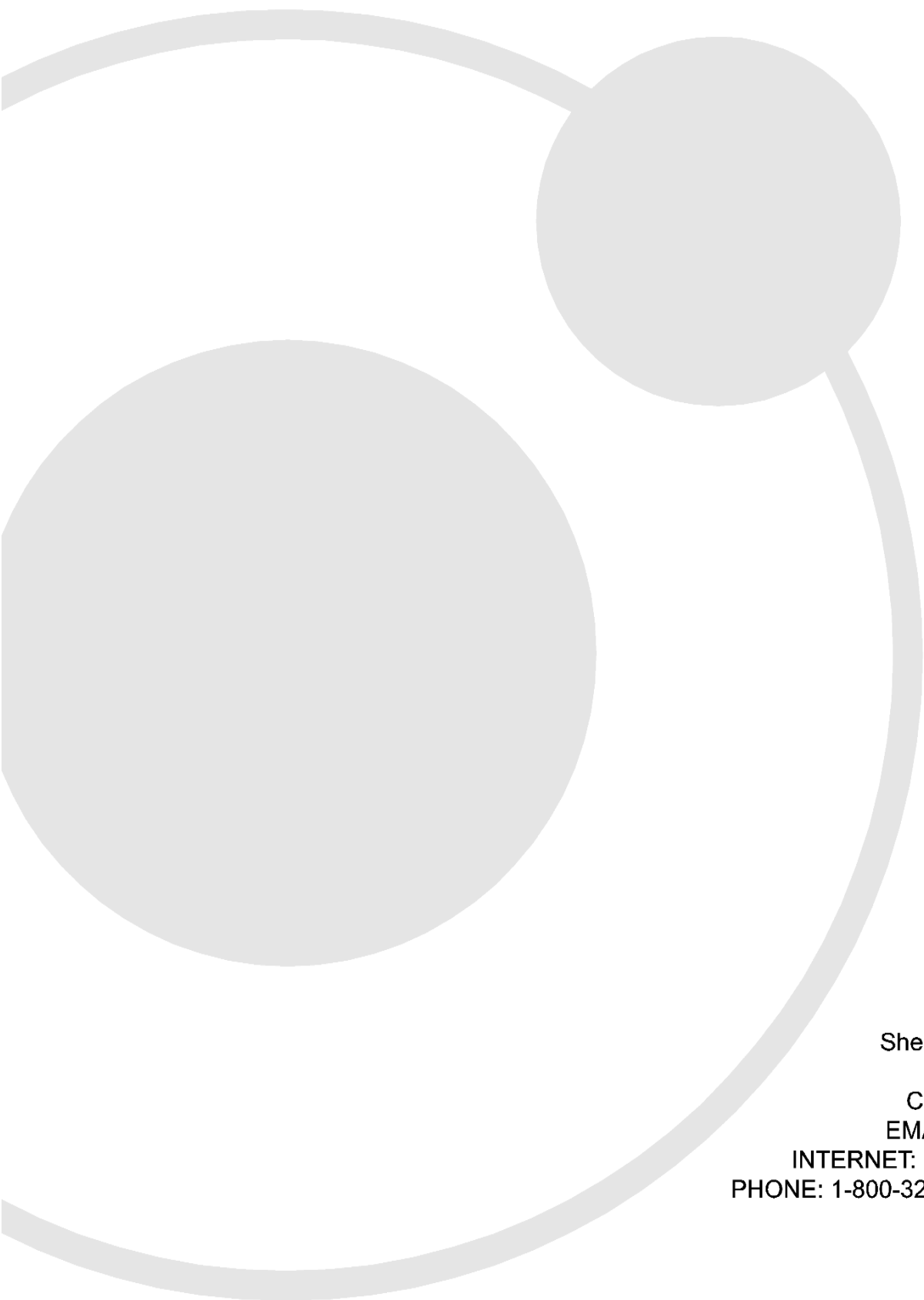
¹ Courtesy of NOAA (http://www.srh.noaa.gov/epz/?n=wxcalc_vaporpressure)

3) Calculating the O₂ Concentration (%)

$$O_2\% = \left(\frac{PPO_2}{BP} \right) 100$$

Calculating the O₂ concentration requires:

- **PPO₂**
 - The Partial Pressure Oxygen value calculated in step 2 on the previous page.
- **Barometric Pressure (BP) in millibars (mbars)**
 - The same value previously used to calculate the PPO₂ value.



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