

1. EQUIPMENT INSTALLATION



WARNING: Not follow indications described in this section may cause that the equipment fails or may damage the user.

A. Place the equipment in a flat surface and let at least 10 cm of free space between equipment rear panel and the wall. Never place the equipment in zones with vibration or with direct sun light.



B. Check that AC voltage in the electrical network is the same that the voltage selected in the equipment. Never connect the equipment to a power outlet with voltage out of these limits.



For electrical safety reasons you only can connect equipment to a power

outlet provided with earth This equipment can be used in installations with category II of over-voltage according General Safety Rules.

The manufacturer does not accept responsibility by wrong use of the equipment and the consequences of different use to which it has been designed for.

2. MAINTENANCE



WARNING: Not follow any of the indications on this section may produce wrong working of the equipment.

- PRESS KEYS SOFTLY It's enough to press lightly.
- Clean the equipment with a dry piece of robe or a lightly moistened one. NEVER USE NEITHER ABRASIVE PRODUCTS NOR DISSOLVENTS
- NEVER pour water or liquids on the equipment.
- Once you have finished using the equipment turn it off by using main switch, clean and check the equipment so that it is in optimal conditions when you use again.
- User is only authorised to replace fuses by the specified type if it was necessary.

BATH POWER SETTING



FIGURE 1. Resistor voltage selector

Before connecting the bath to the thermostat, you must check inder the pump the voltage selector is correctly set (115V or 230V) and change it in case it was necessary. In this way the heating element will have a power of 500W.

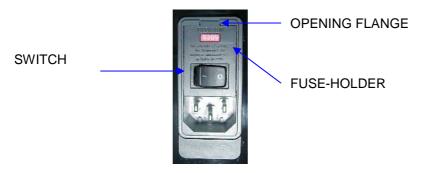


Figure 2. Power inlet, main switch and fuse holder

FUSES REPLACEMENT OR VOLTAGE CHANGE PROCEDURE

In case of an over-voltage or another incidence in the AC net, if it's not possible to turn on the equipment. Or when the AC voltage of the net is different that the voltage of the equipment. Check fuses according the following procedure.

- 1. Remove power cord from the power inlet
- 2. Open fuse-holder pulling the flange with the help of a flat screwdriver



Figure 3. Open fuse-holder door

3. Extract fuse holder

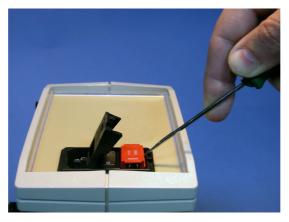


Figure 4. Extract fuse-holder.

- 4. Replace fuses in case it was necessary.
- 5. Insert again fuse holder so that the correct voltage can be read (115V or 230V) and close again the cover
- 6. In case that fuses blew again unplug the equipment and contact with technical service.



For electrical safety, never open the equipment. The power supply has dangerous voltages.

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4. INTRODUCTION:

In a Langendorff experiment an isolated heart is maintained in an organ bath while the coronary arteries are perfused with a buffered, oxygenated, nutrient solution. The experiment can be done under conditions of constant perfusate flow or pressure, and the heart can be paced by an external stimulator.

The perfusate does not flow into the heart in the same manner as blood in the whole animal. Instead, the aorta is cannulated and the solution is delivered in a retrograde fashion down the aorta. The perfusate does not enter the heart chambers themselves but flows through the coronary vascular system, which supplies the heart muscle with the nutrients required for contraction. Part of this solution goes to the right atria through the coronary sinus and can get outside of the organ bath where it can be collected.

5. PARTS OF THE SYSTEM:



- A. Perfusate reservoirs
- B. Peristaltic pump
- C. Coiled tubes
- D. Junction box (heart block)
- E. Three-way valves
- F. Heating rod
- G. Water level sensor

- H. Thermocouple
- I. Gas inlet. Bubbling regulators
- J. Heated jacket for heart
- K. Diffuser
- L. Emptying key
- M. Security Thermostat

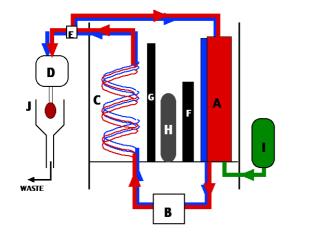
6. CIRCUIT OF THE SALINE SOLUTIONS (PERFUSATES):

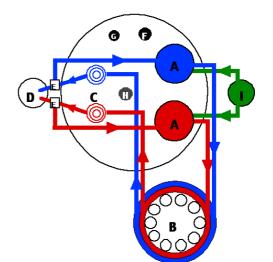
We have two water-heated perfusate reservoirs in which we can have two different nutrient solutions (perfusates). These reservoirs are constantly oxygenated and kept at a constant bath temperature.

The solutions get out of the bath and pass through tubes around the peristaltic pump in order to be pressurised. The pressurised perfusate fluids are warmed again when they re-enter the bath unit in coiled tubes and feel into the junction box (also called heart block) which contains the cannulated heart. Both solutions pass around the pump and towards the junction box, however only one is used at a time to perfusate the heart. The other solution is redirected via the three-way valve and returns to the reservoir. You can alternate between the two different perfusate solutions.

The heart is perfused through the heart block allowing the entrance of the perfusate into the aorta cannula and the access of devices for measuring the temperature and pressure of the perfusate.

The formation of air bubbles is prevented so that no boluses of air enter the heart upon reservoir changes. Additionally the temperature is maintained to prevent cool solution from being perfused into the heart.





7. CONTROL OF THE OXYGENATION:

A bubbling system sends oxygen to the perfusates inside the reservoirs, there are two keys that regulate independently how much bubbling each one of them gets. This system is connected to an oxygen bottle whose output pressure is set at the manoreducer at 0.5 bars.

8. CONTROL OF THE VOLUME OF LIQUID INSIDE THE CONTAINER:

This liquid is distilled water with some fungicide to prevent the growth of seaweed and fungus. You fill the glassware up to a level where the water level sensor and the perfusate reservoirs are covered. This water level sensor is a blue float, which should be always covered by liquid. If this float gets dry, there is an emergency system that disconnects the heating to prevent the burning of the whole glassware. The liquid is constantly circulating. Underneath the glassware, there is a magnetic pump throwing liquid through the diffuser and recovering it through a small whole at the bottom of the glassware. The glassware can be emptied manually by a key.

Some of this liquid leaves the glassware to the water-jacketed organ chamber in order to maintain it at the right temperature, runs all over this small chamber and goes back into the glassware.

9. HOW TO START:

Before placing the heart we should assure that everything is OK. We fill both perfusate reservoirs with two different solutions (or with the same one if we just want to use one solution, then we will have more quantity ready). We regulate the quantity of oxygen going into the reservoirs. Finally we open the three-way valve of the solution we want to use and we close the valve of the other one.

We also fill the glassware with the distilled water with fungicide and we set the temperature we want to achieve. We set the parameters at the peristaltic pump, depending if we want to work in a constant flow or pressure condition.

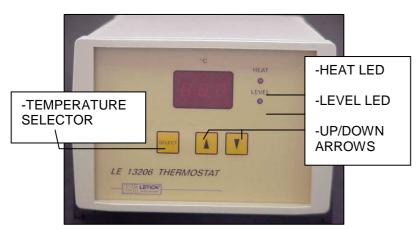
We make everything start and run for several minutes until the right temperature and liquid distribution is achieved.

When we have achieved the right conditions we can place the heart at the heart block and inside the jacketed heart chamber. Two cannulas are provided:

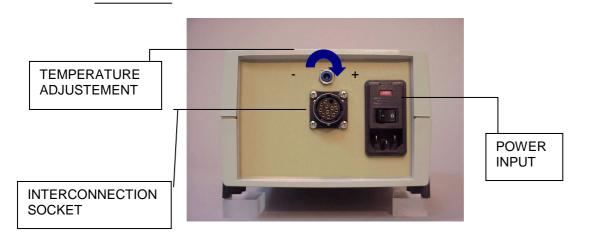
- 2.1mm \emptyset , total length 30mm and 0.7mm \emptyset , total length 30mm

10. THE THERMOSTAT UNIT LE 13206

10.1. FRONT PANEL



10.2. REAR PANEL



10.3. **DESCRIPTION**

The Thermostat Unit LE 13206 controls the temperature of the water in the Langendorff container from room temperature up to 45 °C. It supplies energy to the heating rod of the container and constantly measures the temperature, showing its current value on the display. Temperature is taken from a thermocouple in the center of the container base. All the system is protected by a security thermostat near the resistance.

The Temperature adjustement is used to adjust finely the bath temperature when the HEAT led is off. To increase the temperature turn in to the right very carefully and slowly to see the changes.

In order to achieve a correct propagation of the heat of the thermostatized water, a highly efficient magnetic pump is used. The pump draws the water in by an opening in the container's base and discharges it by the diffuser. This produces perfect heat

diffusion throughout the container, with no visible movement of water and minimum vibrations.

It is important to point out that the thermostatized water must never reach a temperature higher than 45 °C in order to prevent damage to the container and the consequent liquid loss.

The heating rod has an oversized power potential to facilitate rapid heating and to ensure that the set temperature is reached in a short time and with minimum fluctuations.

The heating rod is controlled by means of an original electronic circuit in the Thermostat Unit. This, through an electric feedback mechanism, supplies an energy proportional to the difference between current and target temperatures.

The greater the difference between target and current temperatures, the greater the heat supplied by the heating resistor. Heating power is slowly reduced as soon as current temperature approaches the target one. In this way, temperature is kept smoothly on its target value, without fluctuations or energy peaks. Regulating margins of less than 0.1 degrees throughout the entire volume of water in the container are achieved.

Sometimes there is a small difference (0.3°C) between the temperature we fix and the one we get in the jacketed heart chamber (lower).

10.4. OPERATING INSTRUCTIONS:

The following are the steps to be completed in order to start up with the LE 13206 Thermoregulator:

- 1. Be sure the container is full of distilled water.
- 2. Push the ON/OFF switch of the Thermostat Unit to its ON position.
- 3. The LEVEL LED will light when the level of the water is the one needed to start the experiment. Before that moment arrives, the heating rod will not work, neither the LE13206.
- 4. Press TEMPERATURE SELECTOR, and introduce by the UP/DOWN ARROWS the target temperature (always between 25 and 45 ° C). If you don't press any key during 15 sec the system begins heating automatically.
- 5. Pulse again TEMPERATURE SELECTOR button, and the appliance will start working until the target temperature is achieved. At this moment, the HEAT LED will stop lighting (it only lights when it is working).
- 6. Wait until display temperature stabilizes. While the appliance gets warm until the desired temperature is achieved, the display will show "------", and the selected temperature when it is achieved.

11. MAINTENACE:

In order to longer the Langendorff life, it is convenient to full clean the circuit of the solution periodically. It is also advisable to clean the container when necessary, as indicated. Avoid all abrasive surfaces as scours. Most of the pieces can be removed easily and all the connections are Luer Lock.

Once the perfusate reservoirs and the container have been emptied (it is not necessary to remove them from their place in the container) proceed with the cleaning as follows:

- 1. Make sure that the bubbling regulator is closed. Rub the inside of each reservoir with a brush soaked in soapy (if possible foam less) solution.
- 2. Introduce a slightly acid solution (0.25% sulfuric acid or 0.1 N Hydrochloric acid) into the vessels and supplementary pipes, leaving it to rest for about 30 seconds.
- 3. Immediately after using the above solution, rinse with a large amount of tap water through all the equipment.
- 4. Finally, rinse with distilled water. Check that the bubbling system is working all right again.

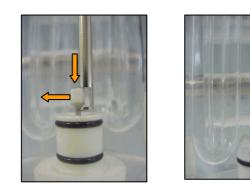
There may exist other ways of cleaning, such as the use of chromic acid or other products. The election is left to the experimenter's judgement.

In spite that the materials being used are resistant to chemical agents, it is also recommended to take care with the taken choices (kind and concentration).

Never use alcohol derived products to clean plastic parts, because this will produce small cracks in the plastic that may cause breaking with time. To clean plastic parts just use a soapy solution and then clean with distilled water.

12. REMOVING AND CHANGING PIECES: 12.1. NEEDLE-VALVE:

□ Take the NEEDLE-VALVE CHANGER, and introduce it inside the vessel. Using the part with a cleft on it, catch the needle-valve and take it out, pulling softly (reverse way to put another).



USER MANUAL

