



UNO-COMBINED-CONTROLLER

CO₂ Microscope Electric Top Stage Incubation System

Manual

Vers. 03.16

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Index

1	REVISION TABLE	5
2	PREFACE	6
3	SYMBOLS DESCRIPTIONS	7
3.1.1	Symbols in this manual and on the product:	7
3.1.2	Symbols on the product label:	7
4	SAFETY NOTES	7
5	AC SECTION WIRING DIAGRAM	9
6	UNO-COMBINED-CONTROLLER. DESCRIPTION.	11
7	EQUIPMENT AND CONNECTIONS	12
7.1	Equipment Supplied.....	12
7.2	Equipment Required (<i>Not supplied</i>)	13
8	UNO-COMBINED-CONTROLLER. OVERVIEW	13
8.1	Temperature Regulation	13
8.2	CO ₂ Regulation	13
8.3	Humidity Regulation.....	14
9	UNO-COMBINED-CONTROLLER. INSTALLATION AND USER GUIDE.....	15
9.1	Digital Temperature Controller connection ports.	15
9.2	Tubing Connections.....	16
9.3	Pressure Gauge set up and use	19
9.4	Assembling Instruction of the Bubbling Column	21
9.5	Cables connection.....	21
9.6	Incubation chamber	24
9.7	Fine gauge thermocouple (Free Sensor-optional)	27
10	CONTROL UNIT FRONT PANEL.....	28
11	TEMPERATURE CONTROL MECHANISM IN STANDARD CONFIGURATION	29
12	TEMPERATURE CONTROL MECHANISM IN CUSTOM CONFIGURATION AT 37.0°C	31
13	UNIT OPERATION THROUGH PUSH BUTTON INTERFACE.....	34
13.1	Main Page, Main Menu And Submenu Description	34
13.2	Button functions	35
13.3	Main Page and Main Menu Navigation	36
13.4	Changing Set Points	37
13.5	Selecting Offset Mode	37
13.5.1	Standard Offset Mode	37
13.5.2	Custom Offset Mode	40
13.6	Objective Heater Submenu	41
13.7	Humidity Module Submenu	44
13.8	Alarm Settings Submenu.....	45

13.9	Screen Settings Submenu	46
13.10	Product Information Submenu	47
13.11	Stop Mode.....	47
13.12	UNO-TS software Active Message	48
14	OBJECTIVE HEATER MANUAL CALIBRATION.....	48
15	FLOW METER REGULATION.....	49
16	MAINTENANCE	51
17	SUPPORT	52
17.1	Web Conference For Assistance And Training	52
17.2	Troubleshooting	53
17.3	Technical Support	54
17.4	ItalyTechnical Specifications	54
18	FIGURES.....	55

1 Revision table

Revision Number	Additions or changes	Date	Editor
01	Edited	November 2014	Amelia Liccardo
02	Review	March 2015	Silvia Foppiano Amelia Liccardo
03	Minor Review	June 2016	Francesco Marino

2 Preface

UNO-COMBINED-CONTROLLER together with a stage top Electric chamber (preventing CO₂ of the desired concentration from dissipating away from the sample) is a compact solution for long term time lapse imaging. We will refer to UNO-COMBINED-CONTROLLER as 'UNO' from here on. 'UNO' is designed to maintain all the required thermal and gas conditions for keeping cells alive for the duration of the time lapse imaging session. "UNO" has a temperature control range of 25-50°C. The system includes a Temperature Control Unit, a manual Gas Mixer -with floating ball flow meters-, and a passive Humidity Module with its own heater. The UNO-COMBINED-CONTROLLER can be connected to a stage top Electric Environmental Chamber from here on referred to as '**Incubator**'. As *OPTIONS* a fine gauge thermocouple can be added to monitor the temperature of a Reference Well located in close proximity to the experimental one. An Objective Heater is *REQUIRED* when using oil immersion objectives. Additionally, UNO-COMBINED-CONTROLLER can be equipped with an optional air pump: OKO-AP, which is a plug and play solution for Air inlet and a convenient alternative to 100% Air tanks/compressed Air lines. All the heated components of the 'Incubator' are heated by 24V DC and their temperatures monitored by High-Accuracy thermistors. Temperature sensors with 0.1°C accuracy are embedded into the following components: lid and base of the 'Incubator', humidity module and objective-heating collar. A single thermal controller regulates the temperature of the individual components above in a coordinated fashion avoiding overshooting the target temperature. Each unit undergoes a careful factory calibration guaranteeing that the sample temperature, inside the chamber, is kept at the desired value within the range 25 – 50 °C. The system is equipped with visual and sound alarm alert in case the temperature limit is reached. The unit shows current and set point temperature in the same display. 'UNO' includes an integrated manual CO₂ mixer allowing regulating Carbon Dioxide Concentration in Air within the range 1 – 20%. Before use please read this manual and familiarize yourself with the functions and the operation of the device.

3 Symbols Descriptions

3.1.1 Symbols in this manual and on the product:

The following symbols identify important information to note:



CAUTION or WARNING: This symbol warns you about the risk of electrical shock.



CAUTION or WARNING or IMPORTANT: This symbol warns you of circumstances or practices that can affect the instrument's functionality and must refer to accompanying documents.

Tip ► Supplies you with helpful suggestions.

Note ► Supplies you with important information to successfully setup and use the instrument.

3.1.2 Symbols on the product label:



CE MARKING: This symbol indicates a product's compliance with EU legislation.



PRODUCT DISPOSAL: This symbol indicates that this product must not be disposed as urban solid waste.

IP 30

This symbol indicates the protection degree against ingress of solids or liquids inside the product.

4 Safety Notes

Before operating the equipment please read carefully the instructions and the safety notes. If you have any questions, please contact Okolab.

The equipment must only be used as intended and as described in this Manual.

Technically qualified personnel should only operate equipment.

Do not start up the equipment if some of its parts are damaged.

This instrument is not intended for use in locations where flammable or explosive gases are present.

Handle the equipment with care.

Equipment and its internal parts can be damaged by dropping and by shock.

Do not use a volatile solvent such as paint thinner to clean the instrument.

Deformation or discoloration will occur. Use a soft, dry cloth to remove stains from



the instrument. Not following these instructions can result in damage or breakdown of the device and its accessories.

The products labels can be found on the rear panel of the Main Unit.

Do not exceed voltage indicated in this manual and on the product label.

Avoid excessive induction noise, static electricity, magnetic fields.

Do not expose this instrument to rain or moisture.

Prevent throttling and kinking of cables.

Some equipment parts may reach temperatures above 50°C. Please be careful when handling it.



Check that the cables are well inserted into their connectors so they cannot slip off

This device is not intended for medical use

The display should not be rubbed with abrasive materials or hard objects as it may be damaged.

Power cord of unit should be unplugged from electrical outlet when left unused for long period of time.

VENTILATION, please make sure the vents on the unit are not blocked in any way either partially or fully.

Unit should be situated away from heat sources such as radiators, heat registers, stoves, or other appliances or processes that produce heat

Do not start up the equipment if the supply cable is damaged.

Connect the equipment only to grounded mains power socket.

Do not disconnect cables while in operation.



Do not open the unit. Do not remove cover or back.

Prevent metal fragments or lead wire scraps from falling inside instrument. This may cause electric shock, fire or malfunction.

The unit does not contain user serviceable parts.

Before starting, assemble the equipment while unplugged from an outlet.

Unit should never be used where it can fall or be pushed into water.



International caution symbol marks this device. IMPORTANT: read the “Safety Notes” before installing, using and commissioning this device, as the notes contain important information relating to safety and EMC. Not following these instructions can result in damage or breakdown of the device and its accessories.

We reserve the right to make technical variations.

IN NO EVENT SHALL OKOLAB S.R.L. BE LIABLE FOR ANY DIRECT, INCIDENTAL OR CONSEQUENTIAL DAMAGES OF ANY NATURE, FINANCIAL OR OTHER LOSSES RESULTING FROM ANY DEFECTIVE PRODUCT OR THE USE OF ANY PRODUCT.

5 AC section wiring diagram

Figure 1 shows the alternating current (AC) section, designed to connect internal power supplies (see Figure 1 label C) in parallel to low voltage power grid.

- A. **Power Connector**, OM-BZ0310
- B. **Power supplies**, TDK-Lambda-ZWS150-24 and ZWS5-5.

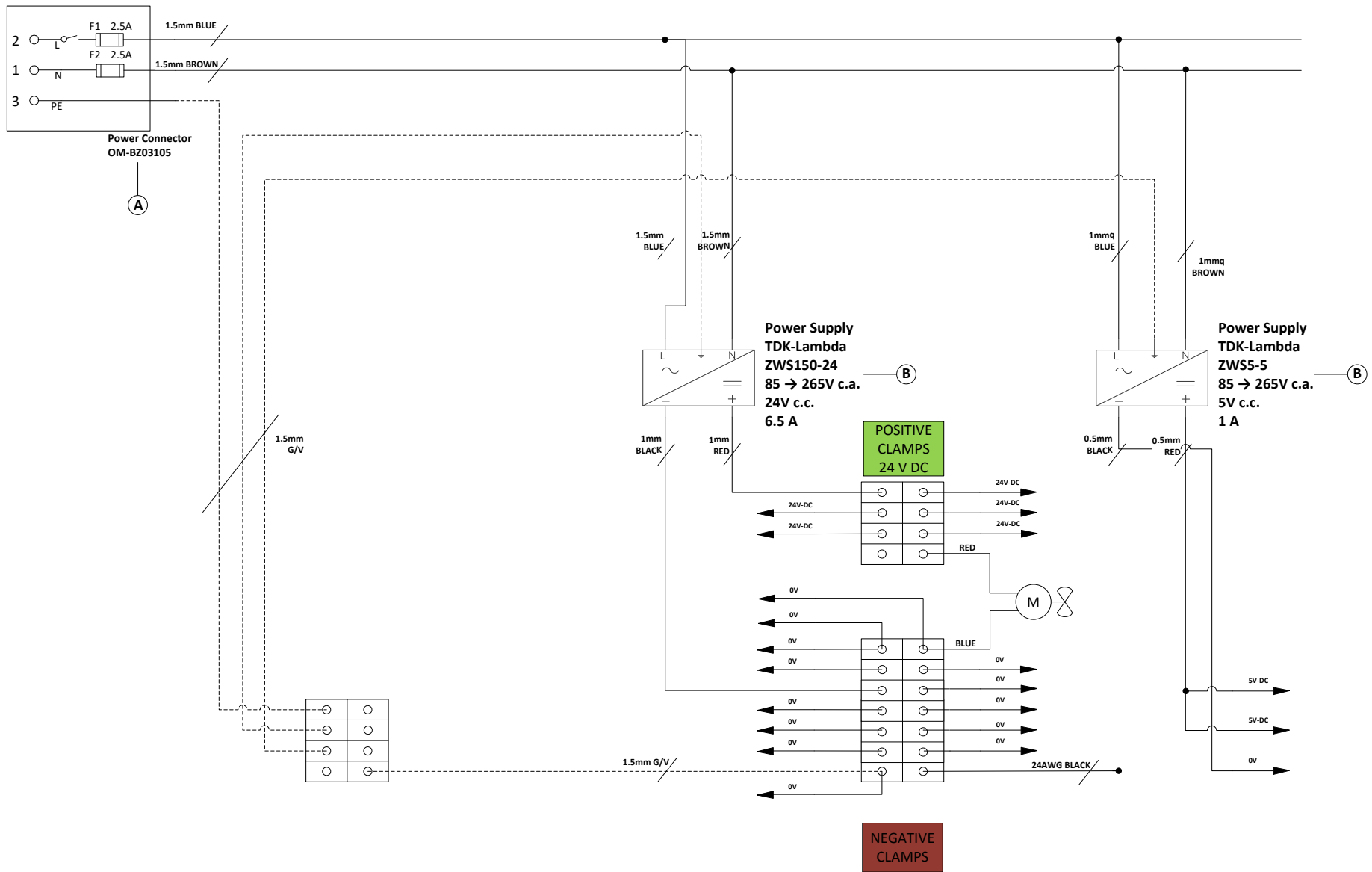


Figure 1 AC section wiring diagram

6 UNO-COMBINED-CONTROLLER. Description.

The **UNO-COMBINED-CONTROLLER** connects Air and CO₂ gas (See paragraph 7).

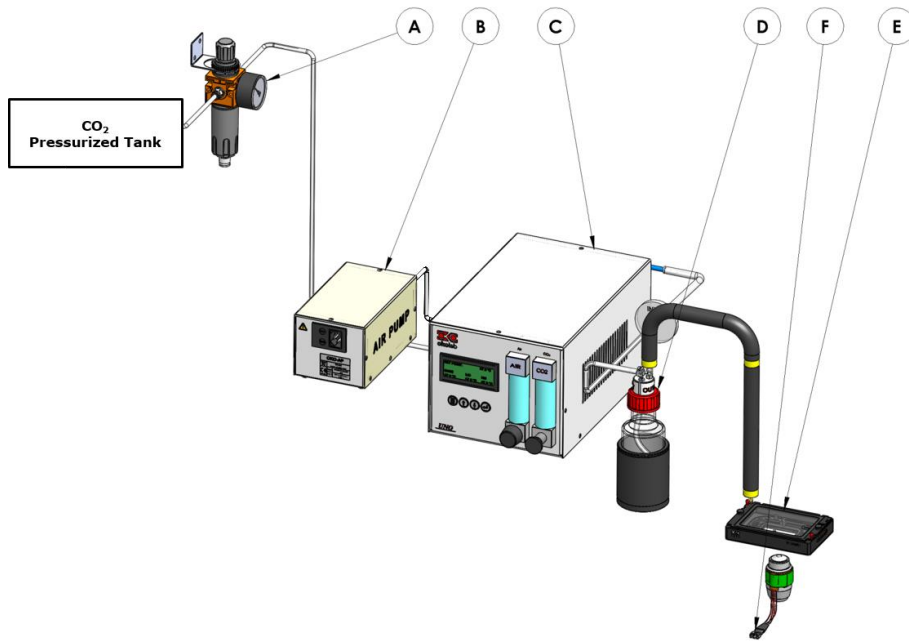


Figure 2. System Overview.

- A. Carbon Dioxide Pressure Gauge with Regulator (Set the pressure at 1bar)
- B. OKO-AP.
- C. Main Control Unit
- D. Humidifying Module with Humidifier Module Heater
- E. Incubating Chamber
- F. Objective Heater (Optional)



When working with Premixed 5% CO₂ remember connecting the rigid tube coming from pressure gauge to Air port labeled "Air In" on rear panel of the Control Unit. In this working modality, Okolab recommends setting a flow rate of 0.3 L/min using the floating ball flow meter on the front Panel of the Control Unit.

7 Equipment And Connections

7.1 Equipment Supplied

UNO includes:

1. **Control-Unit**
2. **Humidifying Module Heater**
3. **Glass Bottle - Humidifying Module** (attention: FRAGILE) **with TUBE E** (140 mm silicon tube of 6 mm OD with Filter).
4. **Pressure gauge with filter, regulator and assembly stirrup for CO₂**, degree of filtration 20µm and condensate drain. Install pressure gauge between the pure CO₂ tank pressure regulator and the Control Unit input port.
5. **Power cord cable (x1)**. Use power cord with Control Unit. The UNO-COMBINED-CONTROLLER can be powered at a voltage within the range 110-230VAC
6. **TUBE E. 3 m long tube assembled as follows:** 2m long rigid tube of 6mm OD, connected to 0.5m long 4mm ID silicon transparent tube. This assembly is then connected to an hydrophobic filter 0.2 micron porous and to a 0.5 m long 4mm ID silicon transparent tube. Connect one end of this tubing assembly to the output port labeled "Air+CO₂ out" and the other end to the Humidity Module.
7. **TUBE H. 1.5 m long tube composed** of 4mm ID silicon transparent tube (0.5 m long) connected to 2mm ID silicon transparent tube (1 m long) (both wrapped in an insulating material). Use insulated tubing in point 8 to connect the Humidity module to the Incubating chamber.

The following items are required but sold separately.

'UNO' must be equipped with:

8. **Incubating chamber** (model depending on the XY stage with or without z piezo) with heated lid, chamber riser and one or more plate adapters.
9. **Flat screwdriver or Allen wrench.** To screw the grub screws or the screws on the sides of the incubating chamber plugging the built in perfusion holes.

Optional accessories:

- **OKO-AP + Power cord (x1)**. The OKO-AP can be powered at a voltage within the range 110-230VAC
- **PRESSURE GAUGE WITH FILTER:** When a compressed air line is available in the lab it is possible to avoid using an air pump such as OKO-AP. In this case a Pressure gauge with filter, regulator and assembly stirrup, degree of filtration 20µm and condensate drain, for Air is available upon request.

Objective heater. Required ONLY when using oil immersion objectives.

- **UNO-TS.** UNO-TS software is available for purchase upon request. UNO-TS includes a Fine Gauge Thermocouple. UNO-TS allows logging temperature data, storing the data in computer memory as well as performing calibrations in the lab. Additionally, the Fine Gauge Thermocouple allows operation in Sample Feedback mode.

7.2 Equipment Required (*Not supplied*)

1. CO₂ gas supply (100%)
2. Compressed Air tank (or a compressed air line). If no air supply is available in the lab please order air pump OKO-AP.
3. Alternatively to 100% CO₂ + air UNO can be operated with a pre-mixed 5% CO₂ gas supply.

8 UNO-COMBINED-CONTROLLER. Overview

UNO-COMBINED-CONTROLLER is at the same time a temperature and a gas controller. It is designed to maintain all the required thermal conditions for live cell imaging and is ideal for time-lapse imaging applications.

8.1 Temperature Regulation

UNO-COMBINED-CONTROLLER controls the sample temperature by regulating the temperature of the base and of the transparent heated lid of the incubating chamber.

Embedded thermistors read the temperature of the incubating chamber's base, chamber's lid and humidifier module. A unique algorithm maintains these temperatures constant. A careful calibration (See Table 1) performed in our laboratories guarantees that sample temperature is maintained at the desired value by selecting the Standard Mode (See paragraph 13.5.1) working modality. An external Fine Gauge Thermocouple (i.e. a thin flexible green wire temperature sensor (*optional*)) can be used to read the temperature of a reference well to calibrate the chamber or to check that sample temperature is at the desired value when using factory default configuration. This optional feature offers higher accuracy of temperature control. Simply place the thermocouple into the reference well and hold it in place with some adhesive tape (See paragraph 13.5). The chambers ('Incubators') accept different plate adapters fitting many kinds of plastic supports, such as 35mm Petri Dishes, chambered slides, simple glass slides and multi-well plates. (See Table 1). Please contact Okolab if you need assistance selecting the correct incubating chamber fitting your microscope stage brand/model and sample adapters.

8.2 CO₂ Regulation

CO₂ is mixed with air in the control unit and is continuously fed into the incubating chamber. You can also use pre-mixed 5% CO₂, connecting the pre-mixed gas supply directly to the Control Unit, to the input labeled 'Air In'.

8.3 Humidity Regulation

Humidifying and pre-heating modules prevent medium evaporation and avoid water condensation on glass and plastic surfaces.

9 UNO-COMBINED-CONTROLLER. Installation And User Guide

The following paragraphs illustrate how to install and use UNO-COMBINED-CONTROLLER.



The following instructions will guide you through a quick installation. For safe operation of the unit please read carefully all the instructions and safety notes.

9.1 Digital Temperature Controller connection ports.

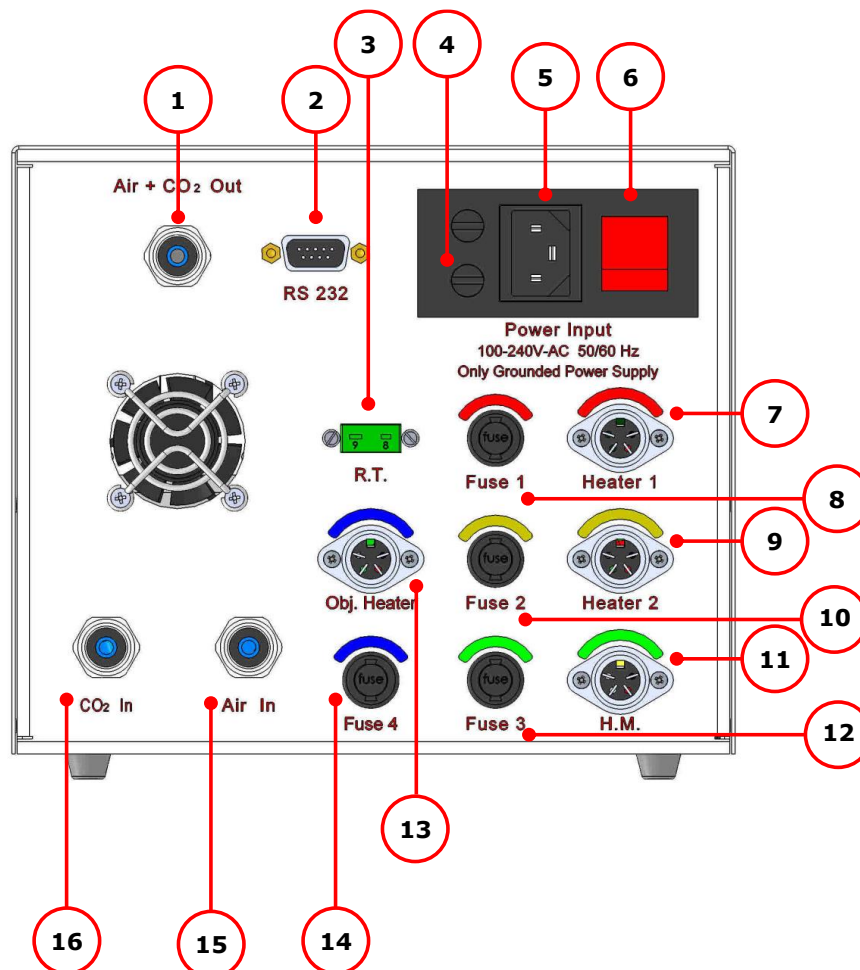


Figure 3. Control Unit Rear Panel Overview

1. CO₂ and Air mixture output
2. RS232 serial port: Software UNO-TS can be used as optional to read incubator temperature and store the data in computer memory
3. R.T.: Fine gauge thermocouple port
4. Time Lag Fuses 5x20 (x2) (see 'Power Consumption' in paragraph 17.4)
5. Power Input
6. Power Switch

7. Heater 1: Chamber Base connector
8. Fuse 1: 1.6 A Fast Fuse
9. Heater 2: Chamber Lid connector
10. Fuse 2: 1.6 A Fast Fuse.
11. H.M.: Humidifying module-heating connector.
12. Fuse 3: 1.6 A Fast Fuse.
13. Obj. Heater: Objective Heater Connector.
14. Fuse 4: 1.6 A Fast Fuse.
15. Pure Air or Premixed Air+5%CO₂ Push to fit Input Connector for 6 mm O.D. rigid tubing
16. Pure CO₂ Push to fit Input Connector for 6 mm O.D. rigid tubing Input

9.2 Tubing Connections.

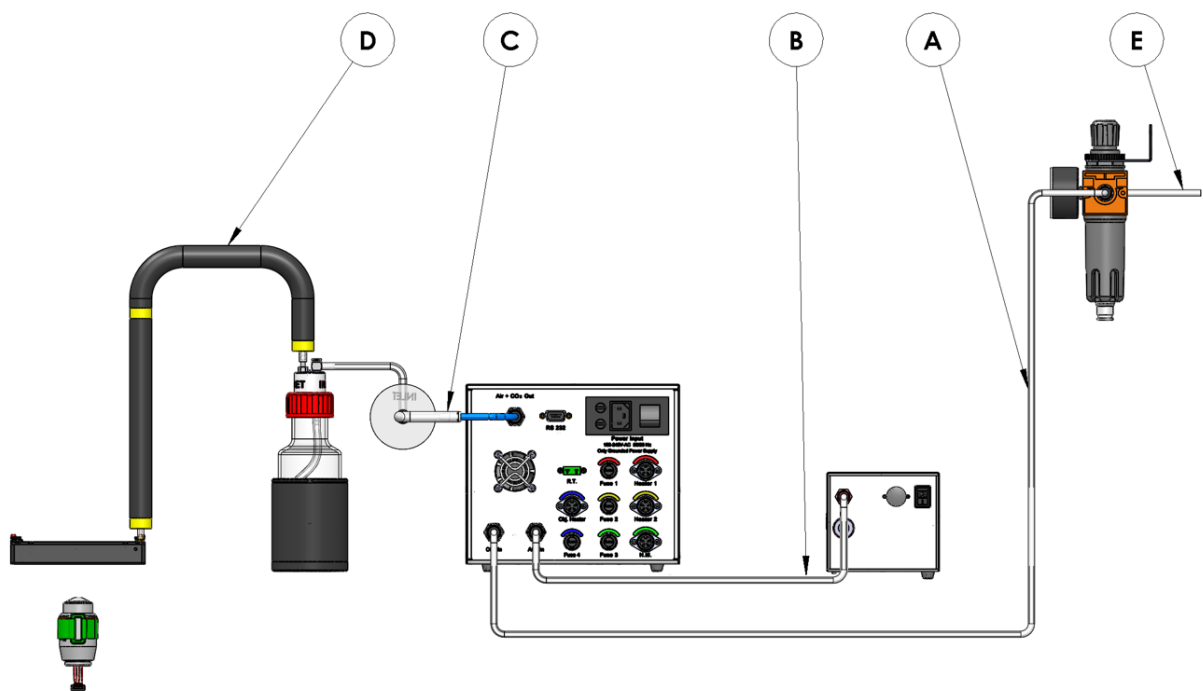


Figure 4. Complete gas lines scheme.

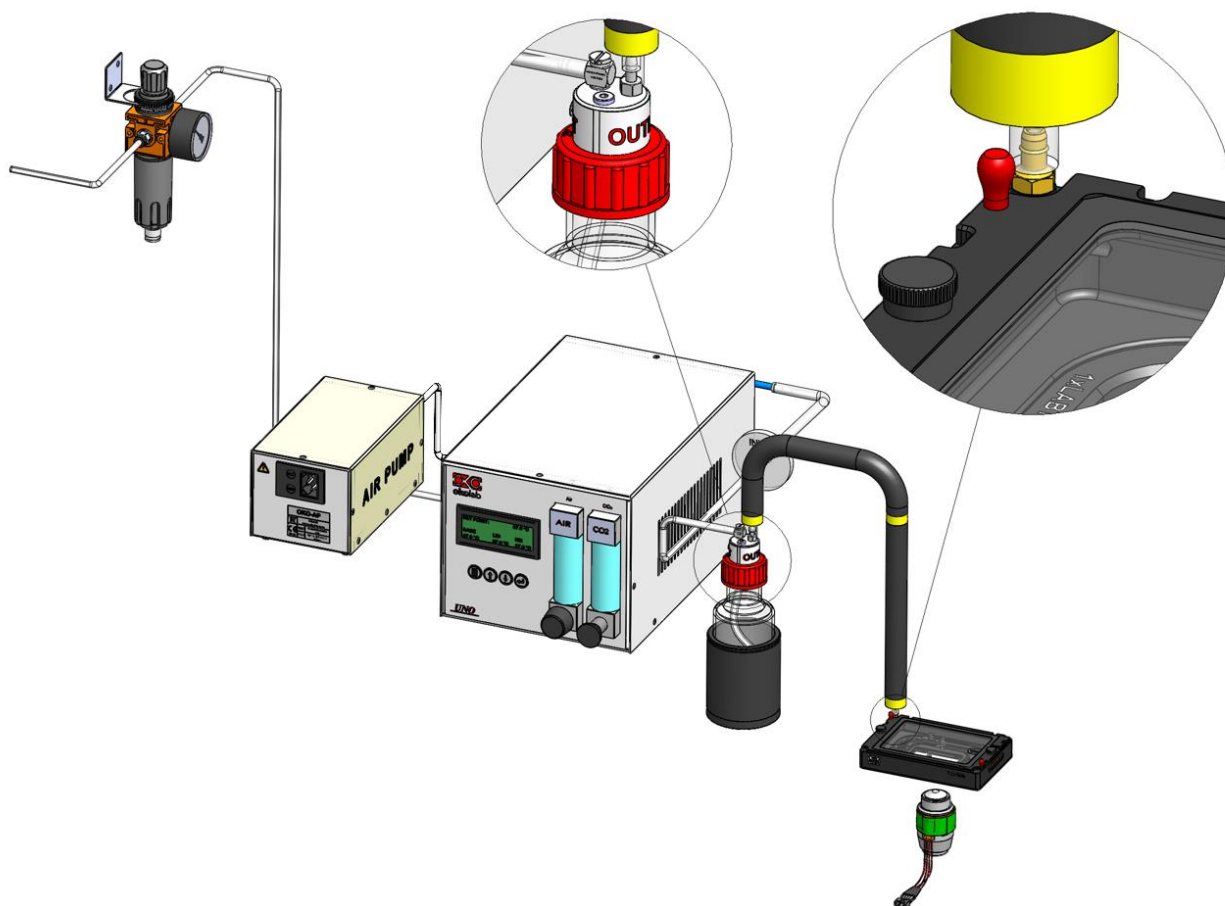


Figure 5. Complete gas lines scheme. Zoom sketches on the Inlet and Outlet connectors of the Humidifier and Zoom sketch on the Inlet connector of the Chamber are reported.

1. Your system requires a pressurized CO₂ tank (not supplied) with safety valve and pressure gauge with regulator within the range 0-6 bar.
2. Connect a rigid tube for pressurized gas (not supplied, see Figure 4), with 6 mm Outer Diameter to the gas output on the pressure regulator of the CO₂ tank. Alternatively fit the pressure regulator of the CO₂ tank with a Swift-Fit Connector for rigid plastic tube with 6mm O.D. and connect TUBE D (supplied, see paragraph 7.1). If you need further assistance please contact Okolab.

Secure the Pressure Gauge for CO₂ with regulator to the wall by using the supplied assembling stirrup (supplied accessories). A filter is added to the Pressure Gauge. Filtering degree is 20µm. CO₂ stream will be FILTERED and at STABLE Pressure. Do not fill the filtering container with oil. Connect the rigid tube 6mm O.D., coming from the pressure regulator of the CO₂ tank (not supplied), to the Inlet of the Pressure Gauge for CO₂ with regulator. Simply push the tubing all the way in. The arrow on the Pressure Gauge with regulator for CO₂ indicates the direction of gas flow as illustrated at Figure 7.

3. Please use the rigid tube, TUBE D (supplied, see paragraph 7.1), 3m long 6mm O.D., (see Figure 4) to connect the controlled (filtered and at P=1bar) flows of CO₂ from the Pressure Gauge to the Control Unit. Connect TUBE D to the inlet located on the back of

the controller and labeled "CO₂ IN". Push the tube all the way in into the Swift-Fit connectors. To remove the tube, pull the tube out while pushing the black ring on the Swift-Fit connector. Follow the arrow on the Pressure Gauge for the correct gas flow direction as illustrated in Figure 7. It is important to ensure that the tubing is properly inserted all the way into the Swift-fit connector in order to avoid gas leaks (See Figure 6).

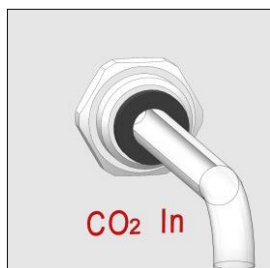


Figure 6. Avoid gas leak: make sure tubing is pushed all the way into the Push-in to-Fit connector

4. Use the rigid tube, TUBE D (supplied, see paragraph 7.1), to connect the Air Output Swift-Fit connector on the front panel of the OKO-AP to the Air Inlet Swift-Fit connector on the rear panel of the Control Unit (Figure 4).
5. Use TUBE E (supplied, see paragraph 7.1) (assembled as follows: 2m long, 6mm O.D. blue rigid tube + to 0.5 m long 4mm I.D. silicon transparent tube + 0.2 micron porous hydrophobic filter + 0.5 m long 4mm ID silicon transparent tube) to connect the Air + CO₂ output Swift-Fit connector on the rear panel of the Control Unit to the Inlet barb connector on the Humidifier bottle, as illustrated in Figure 4 and Figure 5.
6. Use TUBE H (supplied, see paragraph 7.1) (assembled as follows: 0.5 m long 4mm ID silicon transparent tube + 1 m long, 2mm ID silicon transparent tube wrapped in insulating material) to connect the Outlet barb connector on the Humidifier bottle to the miniature barb connector on the Incubating Chamber, as illustrated in the Figure 4 and in Figure 5.

It is possible to use additional filters or filtering devices to further filter the gas mixture prior to its entrance into the incubation chamber further reducing the risk of contamination. If you choose to install an additional filter insert it after the Control Unit and before the hydrophobic filter included in the tubing assembly of TUBE E.

Note ► Use OKO AP when no Air pressurized tank is available.

Note ► Whenever your system is equipped with Premixed gas pressurized tanks with safety valve and pressure gauge with regulator within the range 0-6 bar (not supplied), you can directly connect the Premixed gas to the Air inlet of the Control Unit using Tube D and use the Air regulation in the front panel to control the flow.



Do not remove the hydrophobic filter along the tubing, TUBE C, which connects the Air + CO₂ output Swift-Fit connector on the rear panel of the Control Unit to the Inlet barb connector on the Humidifier bottle. Replace that filter every 6 months. Contact Okolab for further information.

Avoid pinching and kinking of all tubing as it will hinder gas flow and may cause condensation. It is recommended to periodically check tubing connections and gas flow.

9.3 Pressure Gauge set up and use

Follow the direction of arrow on the rear of the Pressure Gauge for the correct Gas In-Out (See Figure 7 below).

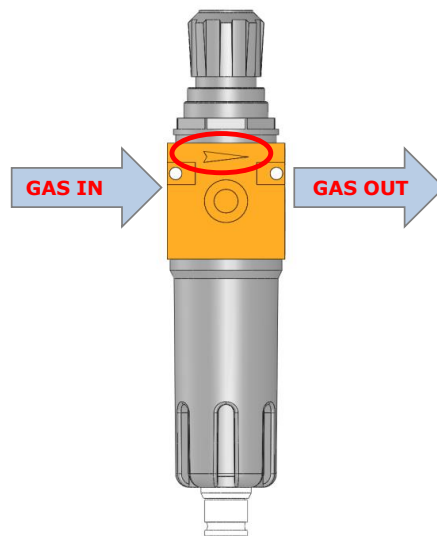


Figure 7. Input and Output gas ports. See symbol on the rear of Pressure Gauge.

On the bottom of the unit there is a purging valve. Close it before operating the Pressure Gauge. To close the purging valve -looking at the regulator vertically from the top knob- rotate it counterclockwise. (See A and B in Figure 8). Now you can operate the Pressure Gauge.

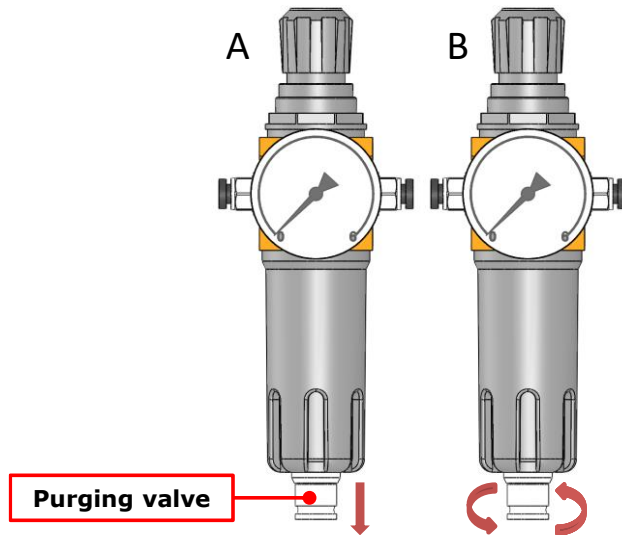


Figure 8. How to close the purging valve of the Pressure Gauge.

To close the gauge, pull the knob up and rotate it counterclockwise. Push the knob down to lock it. Open valve on gas supply tank. Adjust Pressure Gauge by pulling the knob up and rotating it clockwise until the pressure reaches the desired value (See Figure 9).

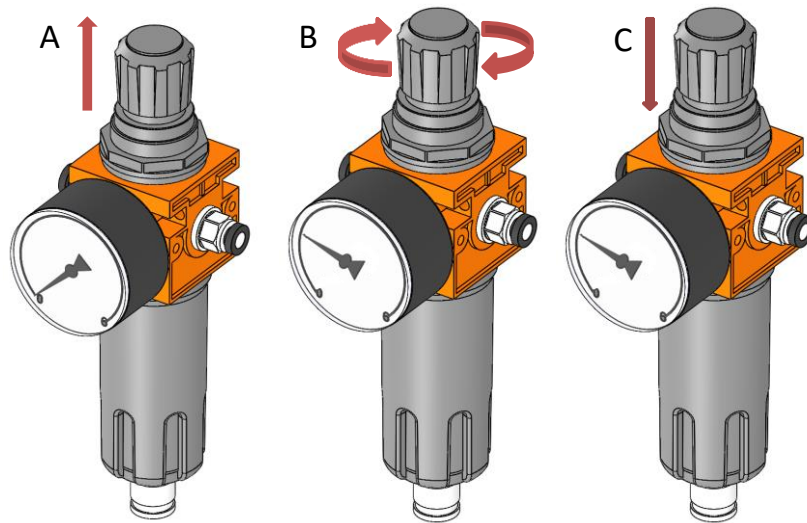


Figure 9. Pressure gauge using.

9.4 Assembling Instruction of the Bubbling Column

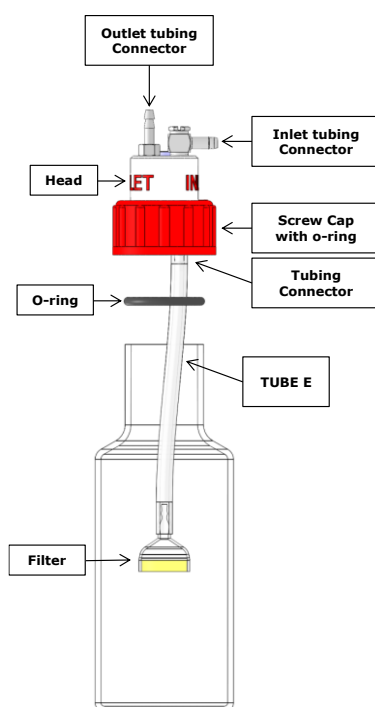


Figure 10. Assembling instruction of the Bubbling Column and its parts

1. Unscrew the red cap with O-ring and the head of the bubbling column from the bottle, without removing the head from the plug.
2. Insert filter into the silicone tubing. Connect the silicone tubing to port of the head of the bubbling column on the inside of the red screw cap.
3. Insert the filter with the tubing (TUBE E in Figure 10) into the glass bottle, place the o-ring on the lip of the bottle neck and then screw the cap on.

9.5 Cables connection.

Refer to Figure 11 in order to connect the cables to Control Unit.

1. Connect the base of the incubation chamber, red labeled, to the port labeled 'Heater 1' on the rear panel of the Control Unit (see Figure 11)
2. Connect the lid of the incubation chamber, yellow labeled, to the port labeled 'Heater 2' on the rear panel of the Control Unit (see Figure 11).
3. Connect the humidifying module heater, green labeled, to the port labeled 'H.M.' on the rear of the Control Unit (see Figure 11).
4. Connect the Objective Heating Collar, blue labeled, to the port labeled "Obj. Heater" on the rear of the Control Unit (see Figure 11).
5. NOTE: you need to connect the Serial Cable to the Serial Port on the rear of the Control Unit ONLY you have purchased the software UNO-TS.
6. NOTE: The optional accessory Fine Gauge Thermocouple (green) (see paragraph 9.7), plugs into the green port labeled 'Reference Temperature' located on the rear panel of

the Control Unit (see Figure 11). Place this thermocouple in a Reference Well filled with water. Make sure that the end of the Fine Gauge Thermocouple is fully immersed in liquid.

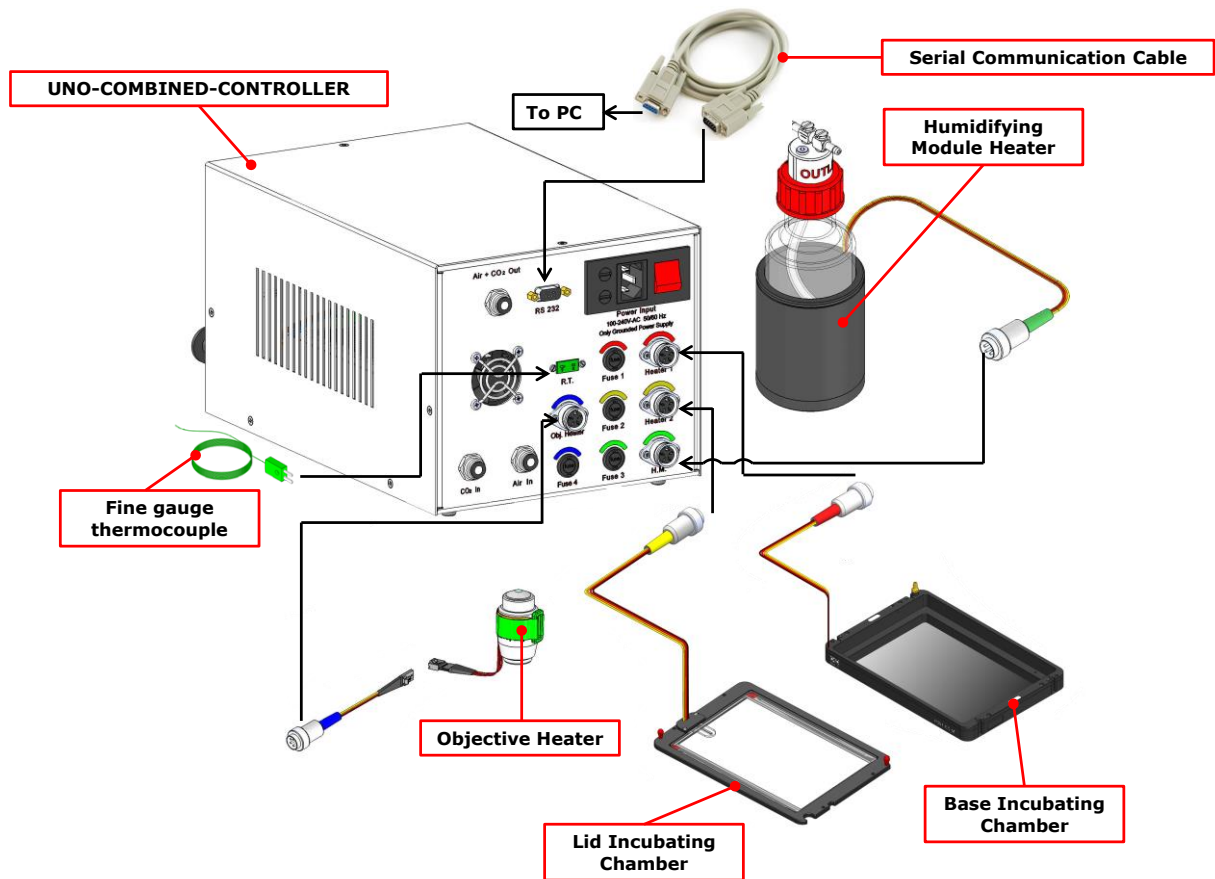


Figure 11. Cables connection. Standard configuration.

- NOTE: an Objective Heater (not included, purchased separately) is required when using oil immersion objectives. The Objective Band Heater kit is designed to fit around any type of microscope objective for upright and inverted scopes. It includes three Heating Bands and three Velcro tapes of different lengths compatible with objectives of different outer diameter and shape as illustrated in Figure 12.

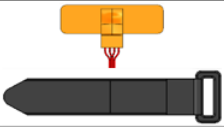
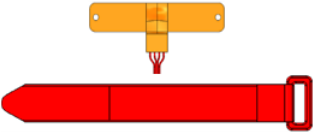
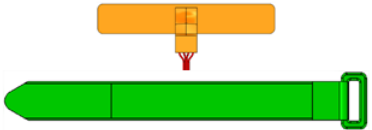
	ORANGEBAND 57mm LONG and BLACK VELCRO TAPE	ORANGEBAND 75mm LONG and RED VELCRO TAPE	ORANGEBAND 98mm LONG and GREEN VELCRO TAPE
LENS' DIAMETER 19-24mm			
LENS' DIAMETER 25-32mm			
LENS' DIAMETER 33-42mm			

Figure 12. Objective Band Heater Selection Chart.

If your system is also equipped with the Objective Heater, (OBJ-COLLAR ####, optional) its objective collar should be installed before connecting the heater to rear panel of the Temperature Control Unit. Follow the steps shown in Figure 13

- Remove the microscope objective and place it on the desk
- Place the orange colored Heating Band around the microscope Objective, cover it with the Velcro tape and fix the Velcro Tape as illustrated in Figure 13
- Then install the Objective to the microscope nosepiece, rotate the Objective carefully avoiding damaging the wires or the flexible heating band.

Avoid holding the Objective Heater Band by lead wires.

Do not modify or cut in any way the Objective Heating Band in the attempt to fit objective of different size.



Make sure that the Objective Heating Band is wrapped around the objective before turning it on. If the Objective Heating Band is on while not wrapped around an objective, it can overheat and can be damaged.

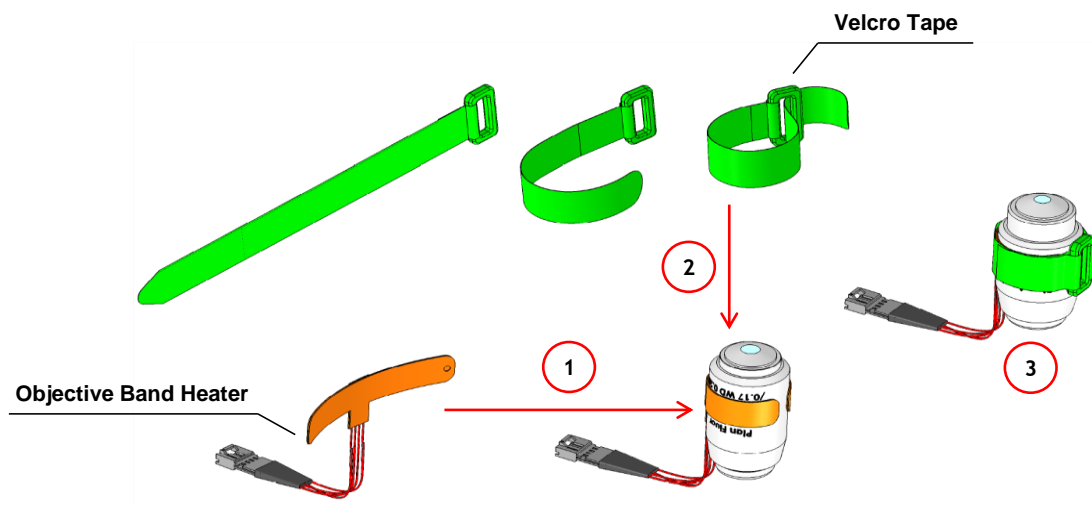


Figure 13. Objective Heater (OBJ-COLLAR #####) Assembly

8. Connect the power cord to the Control Unit.

Once the system has been initialized, the UNO-COMBINED-CONTROLLER display will show its homepage.

Tip ► Wait until the system has reached the steady state before loading your samples.

Tip ► To replace a broken fuse disconnect the unit from the power source. Use a flat head screw driver to replace the fuse if it is broken.

9.6 Incubation chamber

UNO-COMBINED-CONTROLLER must be equipped with an incubation chamber compatible with you stage band/model. Select the chamber on www.oko-lab.com according to your needs. The system is shipped from the factory already set according to the incubating chamber model and sample adapter you have ordered. You can see the current base, lid and HM offsets on the display (See paragraph 13.5.1).

Note ► Thermal control settings are a function of the adapter in use. Hence, every time you change adapter you must input the correct thermal settings using the procedure described in paragraph 13.5.1. In case you purchase additional incubating chamber models make sure to input the correct base offset in the thermal settings prior to starting your experiment. A careful calibration has been performed in our laboratories for each incubation chamber and we guarantee that sample temperature is maintained at the desired value by selecting the adapter and the chamber in use. See Table 1, for values of base offset temperature. Input the correct base offset in the Control Unit Setting using the push button display (See paragraph 13.5.1).

Tip ► The only value that you have to insert, on the push button display, is the base offset, while the lid and HM offsets are automatically calculated when you insert the base offset.

If your sample adapter is not present in Table 1, you can use the Custom configuration and calibrate the chamber in your lab (See paragraph 13.5.2)

Tip ► For a list of all available models and specs of each chamber and adapter see: www.oko-lab.com.

Incubation Chamber	Sample Adapter	Base Offset
H301-K-FRAME H301-NIKON-TI-S-ER H301-OLYMPUS-IX3-SVR H301-PRIOR-H117 H301-NIKON-NZ100/200/500-N H301-MCL-Z100/500 H301-LUDL 96A602 H301-PI-736-160x110	1x35-M; 2x35-M, 2x35-PRIOR-NZ500	-3.3
	2x60-M; 2x60-PRIOR-NZ500	-1.6
	4x35-M; 4x35-PRIOR-NZ500	-1.6
	2xGS-M; 2xGS-PRIOR-NZ500	-4.2
	6MW; 6MW-PRIOR-NZ500	-1.9
	12 MW; 12 MW-PRIOR-NZ500	-2.3
	24 MW; 24 MW-NUNC; 24 MW-PRIOR-NZ500;	-0.6
	24 MW-NUNC -PRIOR-NZ500	
	GS35-M; LABTEK-35-M; LABTEK-II-35-M; LABTEK-35-NZ500; LABTEK-II-35-NZ500; GS35-PRIOR-NZ500	-2.9
H301-NIKON-TI-SR	1x35-M single; 1x60 single	-1.4
H301-OLYMPUS-IX-SUSP	1xGS-M single; 1xLABTEK-M single; 1xLABTEK-II-M single	-2.6
H301-EC-LG-BL	H301-EC-LG-1x35; H301-EC-LG-1xGS; H301-EC-LG-1x60	-3.0
H301-EC-LG-UP-BL	H301-EC-LG-UP-1x35	-2.2
	H301-EC-LG-UP-1x60	-1.9
	H301-EC-LG-UP-1xGS; H301-EC-LG-UP-1xLABTEK; H301-EC-LG-UP-1xLABTEK II	-2.1
H301-UP	UP-1x35-M	-2.2
	UP-1x60-M	-1.9
	UP-1xGS-M; UP-1xLABTEK-M; UP-1xLABTEK-II-M	-2.1
H301-MINI	1x35 single; 1x60 single	-1.5
	1xGS-M single	-2.0
H301-PRIOR-NZ100-H107 H301-PRIOR-NZ100-H117 H301-PI-736-ZR1S/ZR2S		-4.6

Table 1. Chambers and Adapters codes vs Base Offset

9.7 Fine gauge thermocouple (Free Sensor-optional)

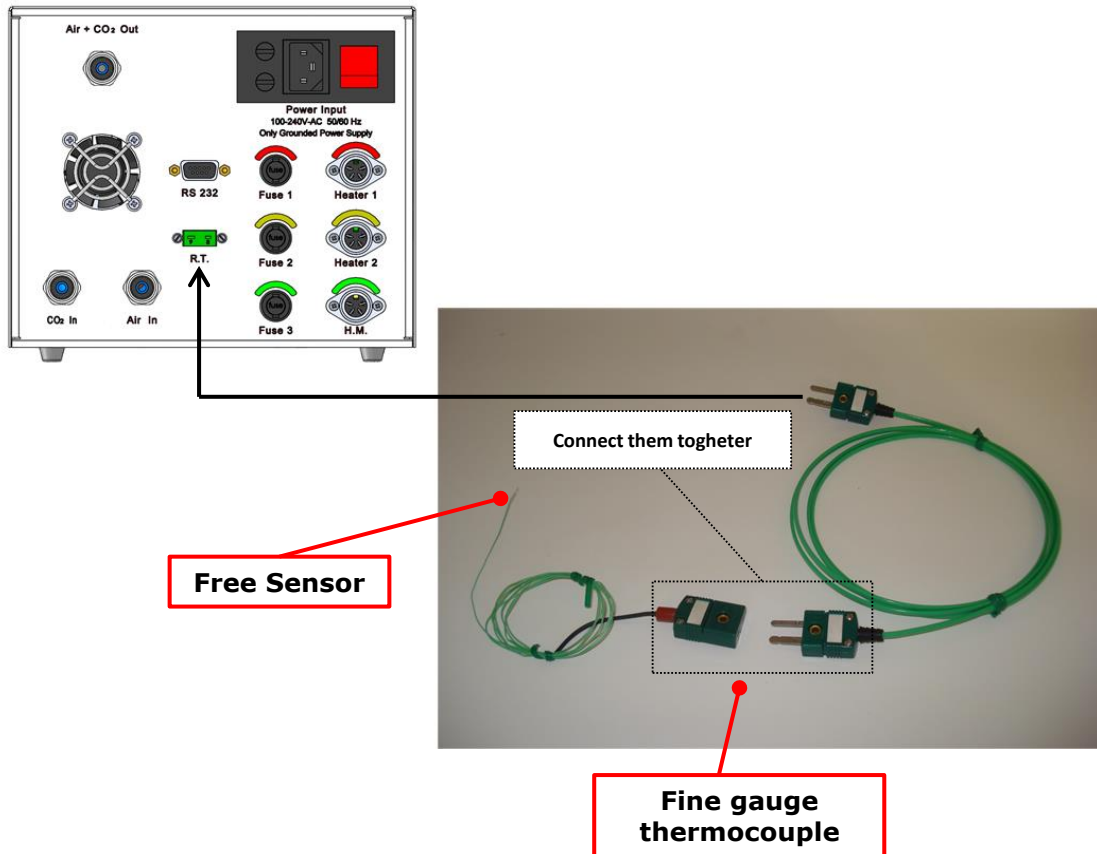


Figure 14. Free Sensor with male to male extending cable.

The incubating system can be equipped with an external Free Sensor (Fine Gauge Thermocouple- Optional added to the system with the purchase of UNO-TS software), which allows measuring the temperature inside a reference well. Use the Fine Gauge Thermocouple to calibrate the chamber's temperature or simply to check the specimen temperature once the steady state is reached.

10 Control Unit Front Panel

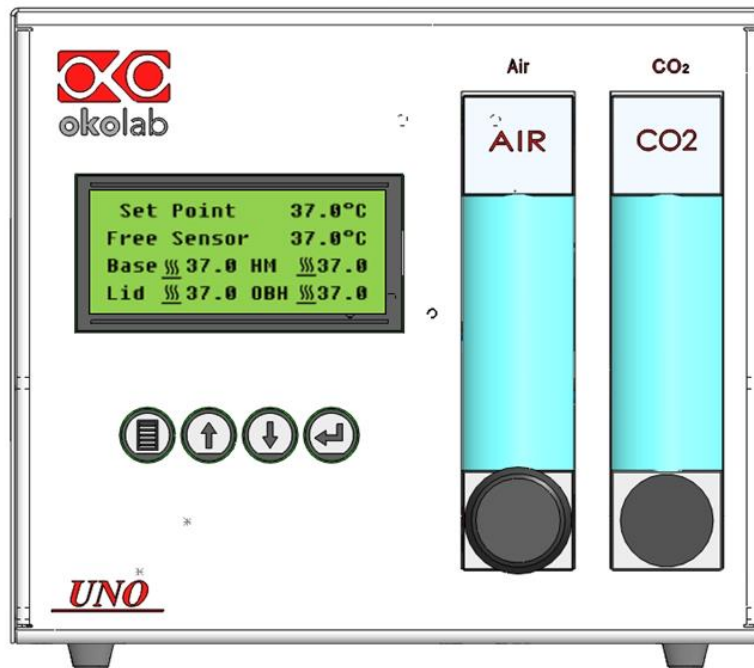


Figure 15. Front Panel of the Control Unit

The front panel of the Control Unit consists of a display with four function keys and a gas mixer. After the unit has been switched on and the warm up screen disappears the display will show the "Main Page". On the "Main Page" the actual chamber's base plate, **BASE**, the actual chamber's lid, **LID**, the Humidity Module, **HM**, and the Objective Heater, **OBH**, temperatures are displayed. The set point temperature is displayed and it can be modified.

1. **BASE** is the actual temperature of the chamber's base plate plus the reading offset.
2. **LID** is the actual temperature of the chamber's transparent heated lid plus the reading offset.
3. **HM** is the actual temperature of the Humidity module plus the reading offset

Tip ► The HM temperature appears only if the HM is active (see paragraph 13.7).


4. **OBH** is the actual temperature of the Objective Heater plus the reading offset.

Tip ► The Objective Heater temperature appears only if the OBH is active (see paragraph 13.6).

5. **Set Point** is the set point temperature.

6. **Free Sensor** is the actual temperature measured by the External Free Sensor (Fine Gauge Thermocouple)

Tip ► The Free Sensor (Fine Gauge Thermocouple) temperature appears only if the free sensor is connected to Control Unit

'Heater on' symbol  : it appears next to each temperature measurement when the heater of each channel is powered on.

11 Temperature control mechanism in standard configuration

The sample, contained in plastic support for cells culture, is maintained at the set point temperature, such as 37.0°C, thanks to a constant metal warming up ensured by the Control Unit.

Tip ► To ensure a sample temperature of 37.0°C, both lid and base should be maintained at a temperature different from 37.0°C. The temperature of the base plate, **BASE**, and of the lid, **LID**, depend primarily on the set point temperature, on the ambient temperature and also on incubator chamber model and on the cells culture support in use (Petri dish, chamber slides or Multiwell plate).

The control display enables the user to calibrate the chamber only selecting the '**Standard**' Mode of the incubation chamber in Submenu '**Offset Mode**' (see paragraph 13.5.1). The control display reads and controls the temperatures of the incubation chamber's parts, adding a reading offset to the **BASE**, **LID** and **HM** temperatures, in order to have the sample at the set point temperature. To show the offsets on the display select **Offset Mode** as 'Standard'. Enter the **Offset** Submenu (see paragraph 13.5.1). Reading Offsets were evaluated with a careful calibration performed by Okolab at ambient temperature of 23+/-1°C. This guarantees that the sample is maintained at the desired set point temperature without the need of further calibrations.

See paragraph 13.2 for buttons functions.

- From the main menu press the 'Menu' key on the instrument panel.
- Press 'Enter' with the cursor on the '**Offset mode**'
- Press 'Enter' with the cursor on the '**Mode**'.
- Press 'Enter' to select '**Standard**'.
- Press 'Menu' Key to exit from the Sub Menu. Press "Menu" key to go to the 'Main Page'.

Tip ► When you select '**Standard Mode**', the row below shows you the base offset value at 37°C.



If you change incubation chamber adapter, you must change the base offset value in order to have the sample at the correct temperature, following the indication shown in paragraph 9.6 and 13.5.1

Once you have selected Standard mode, you can change the set point according to your needs:



- From the 'Main Page', press 'Enter' to change the Set point Temperature.
- Press 'up' or 'down' arrows button to increase or to reduce the Set Point Temperature while it is flashing.
- Press 'Enter' to confirm the new Set Point Temperature.

Tip ► Reading Offsets were evaluated at ambient temperature of $23 \pm 1^\circ\text{C}$ for each Set Point temperature within the working range $25 \div 50^\circ\text{C}$ and stored in the EEPROM memory of the Control Unit. This enables the user to change the Set Point temperature within the working range $25 \div 50^\circ\text{C}$. It also allows maintaining the sample temperature at the Set Point Temperature without the need of any additional calibration.



The Reading Offset values vary with the Set Point temperature and with the chamber and sample adapter in use.

Advantages of this approach:

- The incubating system is Easy to use
- Fast experiment start up 
- Well suited for multi users 
- Sample temperature accuracy of $\pm 0.3^\circ\text{C}$ from the Set Point Temperature.

Disadvantage:

- It needs of calibration in case of the ambient temperature is different from $23^\circ\text{C} \pm 1^\circ\text{C}$.

The default values are optimized for $T_{\text{Ambient}} 23.0 \pm 1.0^\circ\text{C}$ and active Humidifying Module heater.

Offset values are evaluated according to the following:

1. Maintaining sample temperature error within $\pm 0.3^\circ\text{C}$
2. Avoiding condensation on the lid glass
3. Improving the gas stream humidity level

If ambient temperature differs from 23°C of more than 2°C, consider calibrating the chamber as described in the next paragraph.

12 Temperature control mechanism in custom configuration at 37.0°C

The goal is to identify the correct offset (i.e. increase or decrease the controlled base plate and transparent lid temperature manually) of Base and Lid of the chamber, so that the sample cultured in a 35 mm Petri Dish, is at the desired temperature (eg. 37.0 °C). By measuring the sample temperature with the Free Sensor (Fine Gauge Thermocouple), during the steady state, the offset values can be estimated, as described below:

1. Plug the green Fine Gauge Thermocouple in the port labeled "Reference Temperature" located in the rear panel of the Temperature Control Unit, as illustrated in Figure 14.
2. Insert the Fine Gauge Thermocouple into the chamber by threading the wire through one of the perfusion holes on the chamber riser.
3. Secure the end of the Fine Gauge Thermocouple to the bottom of a 35mm or similar Petri dish using some tape making sure to avoid covering the head of the probe (see Figure 16).



Figure 16. Free Sensor attached on the internal base of the Petri Dish.

4. Fill the Petri dish with distilled water to a level sufficient to fully immerse the Fine Gauge Thermocouple. This is a Reference Well simulating experimental conditions. Place the lid on the Petri dish, and insert the dish in the incubation chamber. You can also use SENSOR LID-# to close the dish (see the "**Tip**" below).

Tip ► It is recommended using an Okolab SENSOR LID (available for purchase) to secure the Fine Gauge Thermocouple in place in the Reference Well during calibration (See Figure 17).

Sensor lids are available for the following: 35 mm, 60 mm Petri-dish, chamber slide and chambered cover glass. Sensor Lids are ordered separately using the part numbers in Table 2 below.

SENSOR LID CODES	
SENSOR LID-35	Temperature sensor lid. To be used in specimen feedback in a 35 mm Petri Dish
SENSOR LID-60	Temperature sensor lid. To be used in specimen feedback in a 60 mm Petri Dish
SENSOR LID-GS	Temperature sensor lid. To be used in specimen feedback in chamber slides and chambered cover glass

Table 2. Sensor lid codes.

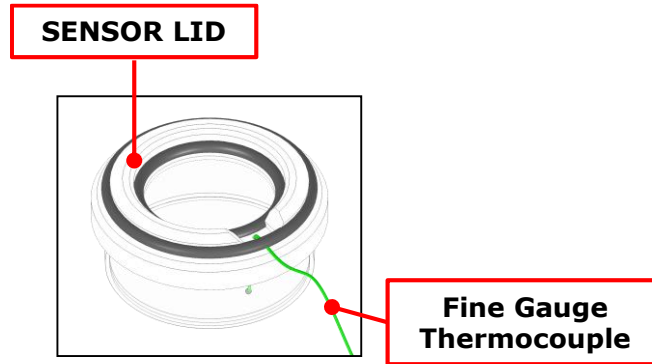


Figure 17. SENSOR LID-35. For 35 mm Petri-dish



It is important to monitor the room temperature during the calibration, as well as reducing air drafts and forced convection. If possible divert any draft coming from the air conditioning system in the room, from the incubation system.

See paragraph 13.5.2 for instructions on how to use the push button display to calibrate the chamber.

- From the main screen press the 'Menu' key on the instrument panel.
- Press move the cursor using the up and down arrows to '**Offset mode**' and press 'Enter'
- Press 'Enter' with the cursor on '**Mode**'
- Press 'Up/down' until you see '**Custom**'
- Press 'Enter' to select '**Custom**' as the configuration in use
- Press the 'Menu' Key to exit from the Sub Menu.
- Press 'Menu' key to go to the 'Main Page'.

Tip ► Reading Offsets can be varied manually to have the specimen at the set point temperature.

Before calibrating the chamber manually, choose the **Set Point** Temperature

- Press 'Enter' to change the Set point temperature to 37.0°C.

- Press 'up' or 'down' arrows keys to increase or to reduce the Set Point Temperature while it is flashing.
- Press 'Enter' to confirm the new **Set Point** temperature.

Go to the Sub Menu '**Offset**' and change the actual Reading Offset of BASE, LID and HM temperatures, in order to have the sample at the set point temperature, as described below. Use table

- From the main menu press the 'Menu' key on the front panel.
- Move the cursor to '**Offset**' with the 'up' and 'down' arrow keys.
- Press 'Enter' with the cursor on the item '**Offset**'.
- Move the cursor to the item '**Base**' using the 'up' and 'down' arrow keys.
- Press 'Enter' to change the Reading Offset of Base temperature.
- Press 'up' or 'down' arrow keys to increase or reduce the Reading Offset of Base temperature while it is flashing.
- Press 'Enter' to confirm the new Reading Offset of Base temperature.
- Move the cursor to the item '**Lid**' with the 'up' and 'down' arrow keys.
- Press 'Enter' to change the Reading Offset of Lid temperature.

Tip ► Set the Offset of the Lid at a value higher than the Offset of the Base by 2°C. Eg. If offset of the Base is +5°C then set offset of Lid to +7°C. As a result the Lid will be at a temperature 2°C higher than the Base. This is done to avoid water condensation on the observation window. With these assumptions the ONLY parameter that needs to be experimentally identified is the Reading Offset of Base.

- Press 'up' or 'down' arrows button to increase or to reduce the Reading Offset of Lid temperature while it is flashing.
- Press 'Enter' to confirm the new Reading Offset of Lid temperature.
- Press 'Menu' Key to exit from the Sub Menu. Press 'Menu' key to go to the Main Page.
- Wait until the steady state of the Free Sensor temperature is reached.
- Increase the Reading Offset of Base and consequently the Reading offset of Lid, if the temperature of the Free Sensor at steady state is lower than 37.0°C.

Table 3 shows Offsets measured at the factory with an insert for a single 35mm dish and a set point of 37°C. Additional Offsets for set points other than 37°C were extrapolated.

Ambient temperature [°C]	Desired well temperature [°C]	Base Offset [°C]	Lid Offset [°C]	H.M. Offset [°C]
23.0 ± 1.0 (Suggested ambient temperature)	32.0	-1.1	-2.2	-1.7
	33.0	-1.1	-2.4	-2.0
	34.0	-1.2	-2.7	-2.2
	35.0	-1.3	-2.9	-2.5
	36.0	-1.4	-3.2	-2.7
	37.0 (Default)	-1.5	-3.5	-3.0
	38.0	-1.7	-3.8	-3.2
	39.0	-1.8	-4.2	-3.5

Table 3: Reading offset estimated value: extrapolated values from trend lines equations. Configuration; 35mm Petri Dish adapter, with Humidifying Module heater. Standard values are pointed out (specimen temperature 37°C.)

Advantages of this approach:

- High accuracy



- Well suited for expert user



Disadvantage:

- Further calibration is needed in case room temperature differs more than +/-2°C from factory calibration temperature of 23°C.
- Offset values are valid only for one Set Point temperature. The Offset values must be experimentally re-evaluated for each different set point.

If the sample adapter changes, please modify the Offset and re-calibrate the chamber, to ensure that the sample will be at the correct temperature.

Offset values are evaluated according to the following:

1. Maintaining sample temperature error within ± 0.3 °C
2. Avoiding condensation on the lid glass
3. Improving the gas stream humidity level by adjusting the offset of **HM**

13 Unit Operation Through Push Button Interface

13.1 Main Page, Main Menu And Submenu Description

```

Set Poi nt  37.0° C
Free Sensor 37.0° C
Base ||| 37.0 HM ||| 37.0
Li d ||| 37.0 CBH ||| 37.0

```



Figure 18. Controller Display "Main Page" and function buttons

The controller has four Function Buttons: "Menu", "up", "down" and "Enter" buttons.

13.2 Button functions



'Menu' button

To access the main menu, press "Menu".

While a parameter is being modified, press "Menu" to escape without saving the parameter.

Use "Menu" button to go up or to exit from a "Sub Menu".



'Up and down' buttons

Press "down" or "up" in order to change the value, when a numerical or logical value is flashing.

Press "down" or "up" to move the cursor along the "Main Menu" items.

Press "down" or "up" to move the cursor along the parameters of a "Sub-Menu".

Press "up" to silence for 30 seconds the audible alarm when active.



'Enter' button

Press "Enter" to have access to a "Sub-Menu" from the "Main Menu".

Press "Enter" after modifying a value to confirm it.

Press "Enter" to change the set point temperature on the "Main Page".

In the Run Mode, press "Menu" and "Enter" at the same time to enable Standby Mode. The controller becomes inactive. Press 'Enter' for re-enabling it.

13.3 Main Page and Main Menu Navigation

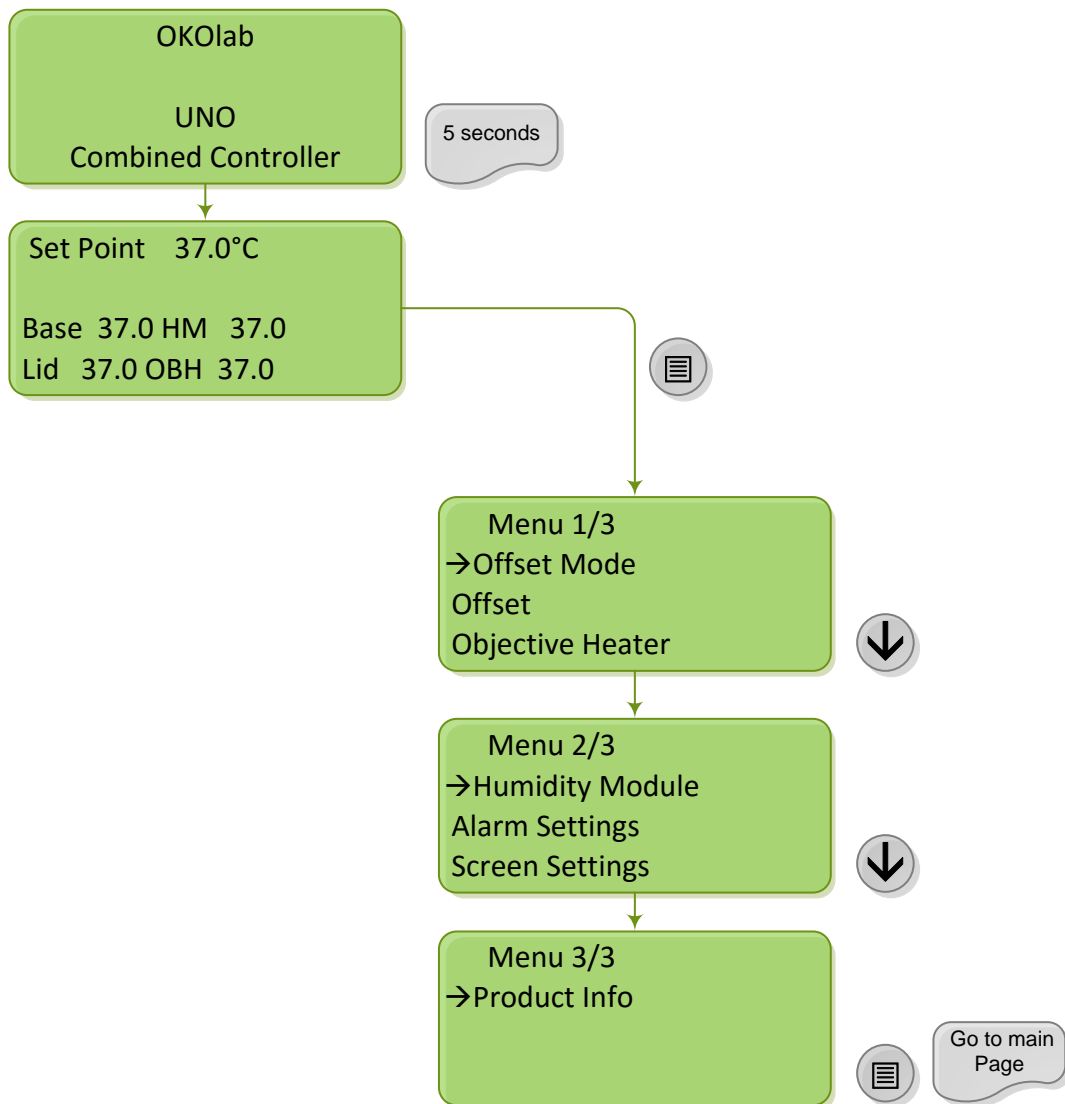


Figure 19. Main Page and Main Menu Navigation Description

To enter the configuration 'Main Menu'

- Press 'Menu' button. Press 'Up' or 'Down' to advance/navigate to the next menu item.
- To Exit from the 'Main Menu' press 'Menu' button

13.4 Changing Set Points

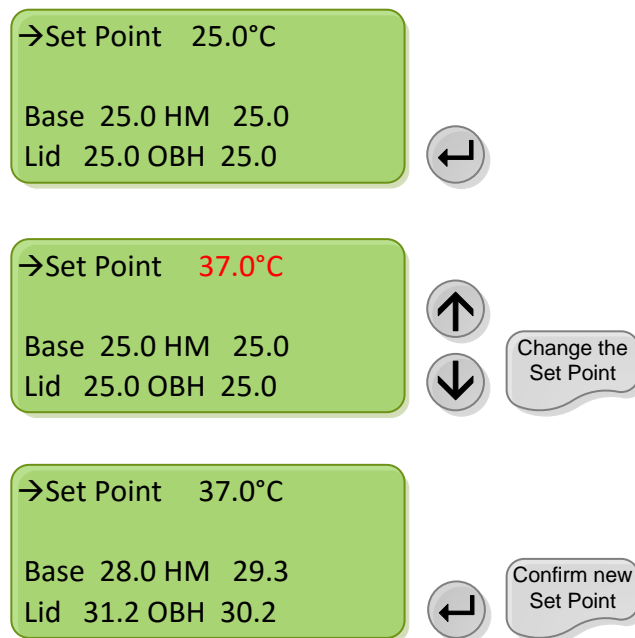


Figure 20. Main Menu Display. Changing Set Points Description.

'Main Page' displays actual temperatures of Base, Lid, Objective Heater and Humidity Module and the Set Point value.

Tip ► The Objective Heater and humidity module temperatures are shown on display only if they are active (See paragraphs 13.6 and 13.7)

- Press 'Enter' to change the set point temperature.

'Main Page' can display temperature of a **Free Sensor** (Optional). **Free Sensor** can be used to monitor the temperature inside the Petri dish, to calibrate the chamber and to check the sample temperature during an experiment.

Figure 21 shows the 'Main Page' when working with Free Sensor connected to the Control Unit.

Set Point	37.0°C
Free Sensor	37.0°C
Base 37.0	HM 37.0
Lid 37.0	OBH 37.0

Figure 21. Main Page when working with Free Sensor connected to the Control Unit.

13.5 Selecting Offset Mode

13.5.1 Standard Offset Mode

When working in standard mode the temperatures of both the lid and the base of the incubating chamber are strictly controlled. A careful calibration performed in our laboratories, guarantees that sample temperature is maintained at the desired set point value.

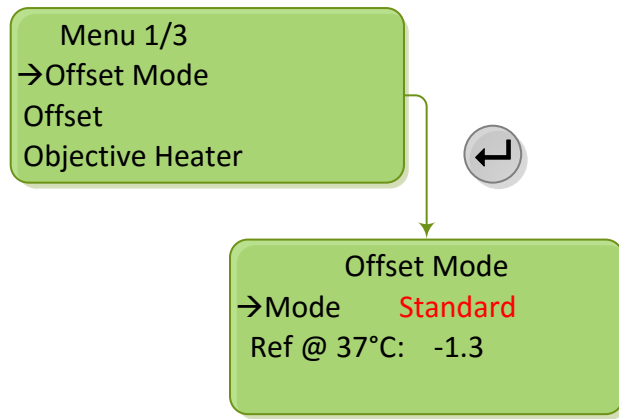


Figure 22. Offset Mode Selecting.

- Enter the Main Menu pressing the 'Menu' Key
- Move the cursor to 'Offset Mode' and press 'Enter'. This will show the current base offset in Standard Working Mode.

Tip ► The system is shipped from the factory already set according to the incubating chamber model and sample adapter you have ordered and in standard working mode. You can read the base offset in use on the display (See Figure 22). You can also see all the current offsets (lid base and HM offsets) in 'Offset' submenu (See Figure 23).

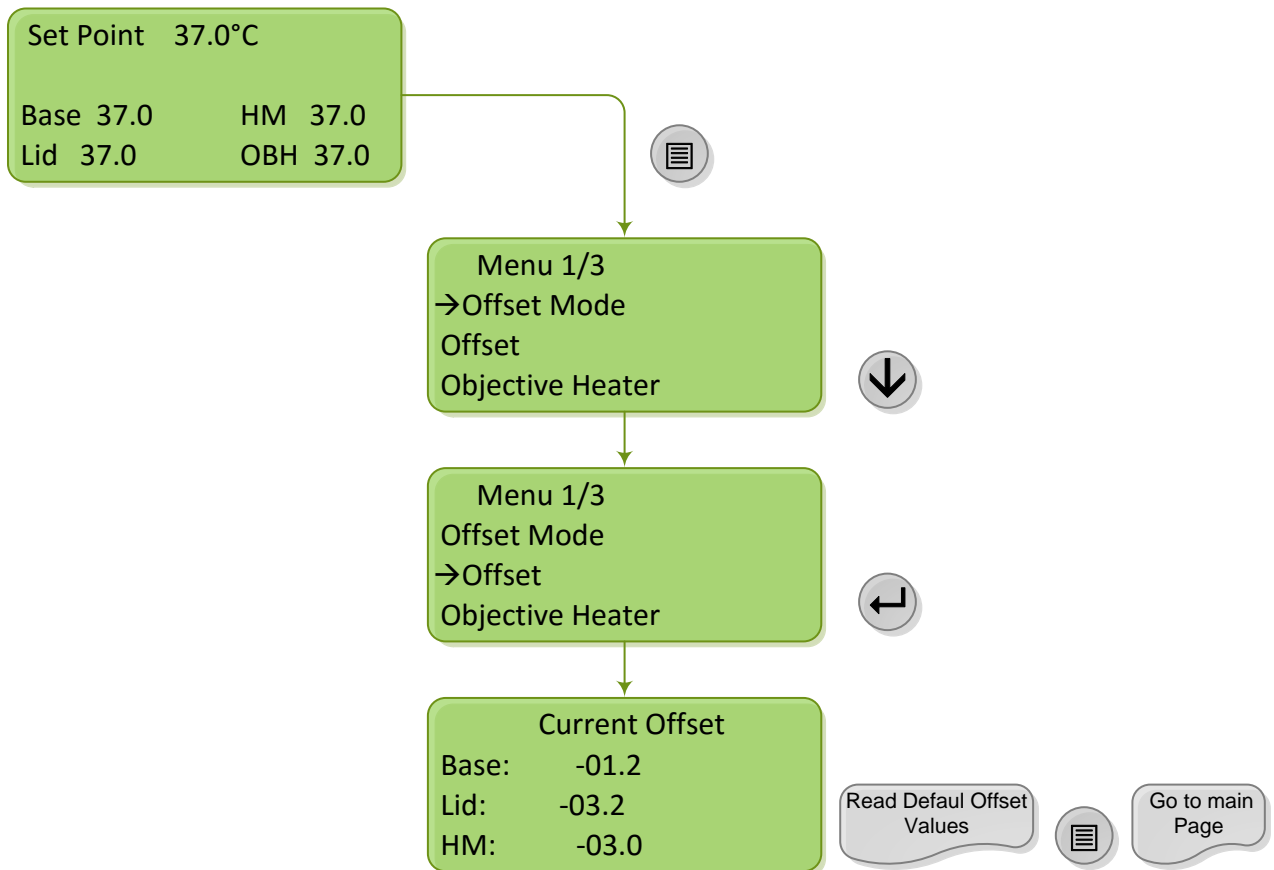


Figure 23. How to see the current offsets in standard mode

Once you have entered the Main Menu pressing the 'Menu' Key:

- Move the cursor to the item 'Offset' with the 'Arrow' Keys and press 'Enter'.

Here you can read the offset values. If you are using a CUSTOM PLATE ADAPTER, not included in the list of standard adapters or if the room temperature differs more than 2°C from 23°C (temperature of the factory calibration) follow this procedure: To change the 'Reading Offset' select 'Custom' configuration. Here you can change the Reading Offset of the base plate, lid and humidity module temperatures in order to have the sample at the set point temperature. Select this configuration only when using a custom plate adapter, or if the room temperature differs more than 2°C (See paragraph 13.5.2)

Tip ► Keep in mind that thermal control settings change when changing the adapter in use. Hence, every time you change adapter you must input the correct base offset using the procedure of Figure 24 and the offset values reported in Table 1. In case you purchase additional incubating chamber models make sure to input the correct base offset prior to your experiment.

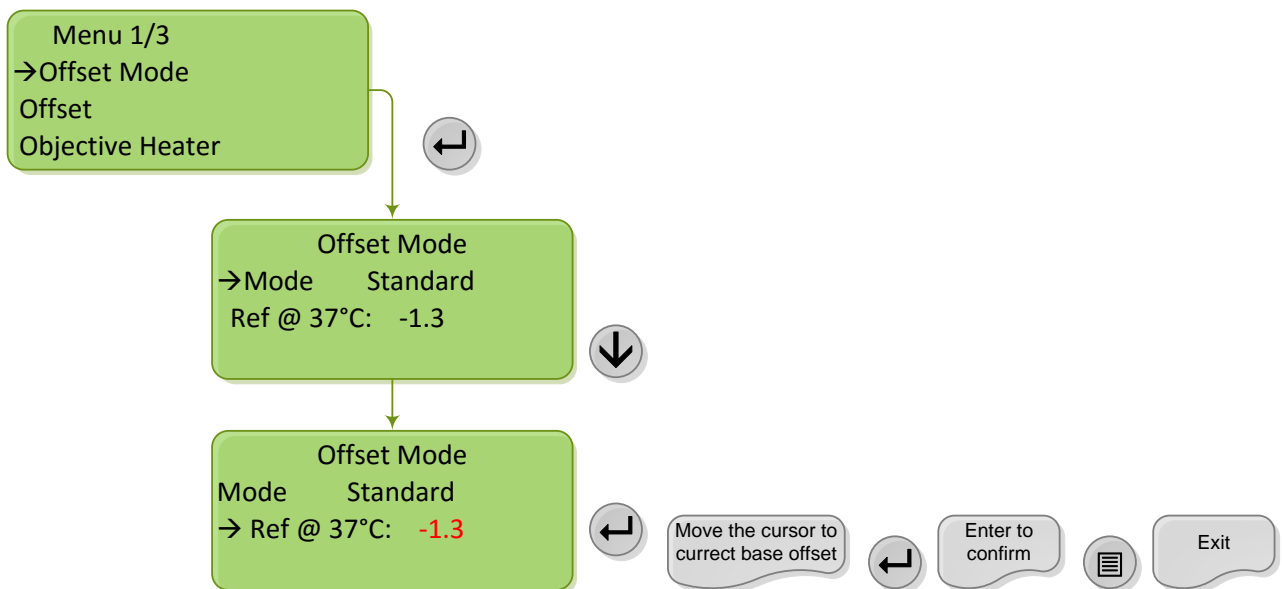


Figure 24. How to change the base offset value.

- Press 'Menu' Key
- Press 'Enter' with the cursor on the item 'Offset Mode'
- Move the cursor to the item 'Ref @ 37°C' and press 'Enter'
- Move the cursor to the correct base offset while it is flashing and press Enter to confirm the chosen offset value.
- Press 'Menu' Key to exit from the Sub Menu. Press 'Menu' key to go to the 'Main Page'.



Please note that the **only** offset value to change is the base offset. Lid and HM offsets will be calculated as a function of the base offset and you will see them in 'Offset' submenu. (See Figure 23).

13.5.2 Custom Offset Mode

In order to select the Custom Offset Mode, see Figure 25. If the room temperature differs more than 2°C from 23°C (temperature of the factory calibration) or if you use a custom plate adapter, non-included in the list reported in Table 1, Okolab recommends this working mode recalibrating the chamber following the procedure reported in paragraph 12.

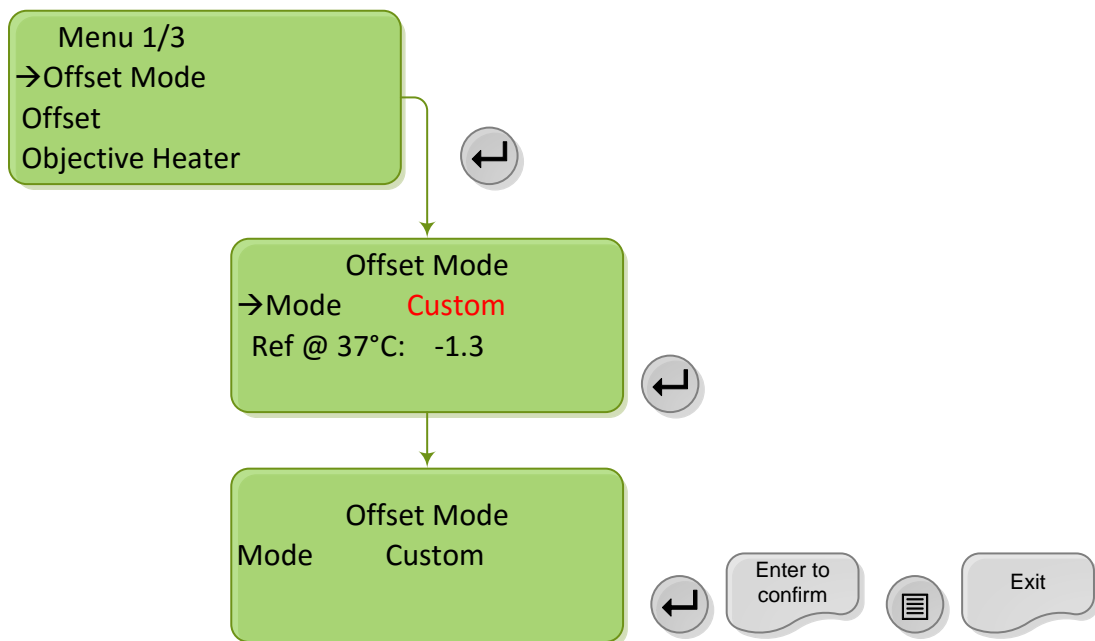


Figure 25. How to choose the Custom Offset Mode

- From the main menu press the 'Menu' key on the front panel.
- Press 'Enter' with the cursor on '**Offset mode**'
- Press 'Enter' with the cursor on '**Mode**'
- Press 'Up/down' until you see '**Custom**'
- Press 'Enter' to select '**Custom**' as the configuration in use.
- Press 'Menu' Key to exit from the Sub Menu.
- Press 'Menu' key to go to the 'Main Page'.

Once you have selected 'Custom' mode, you can change the base, lid and HM offset iterating the same procedure shown in Figure 26 for the three devices.

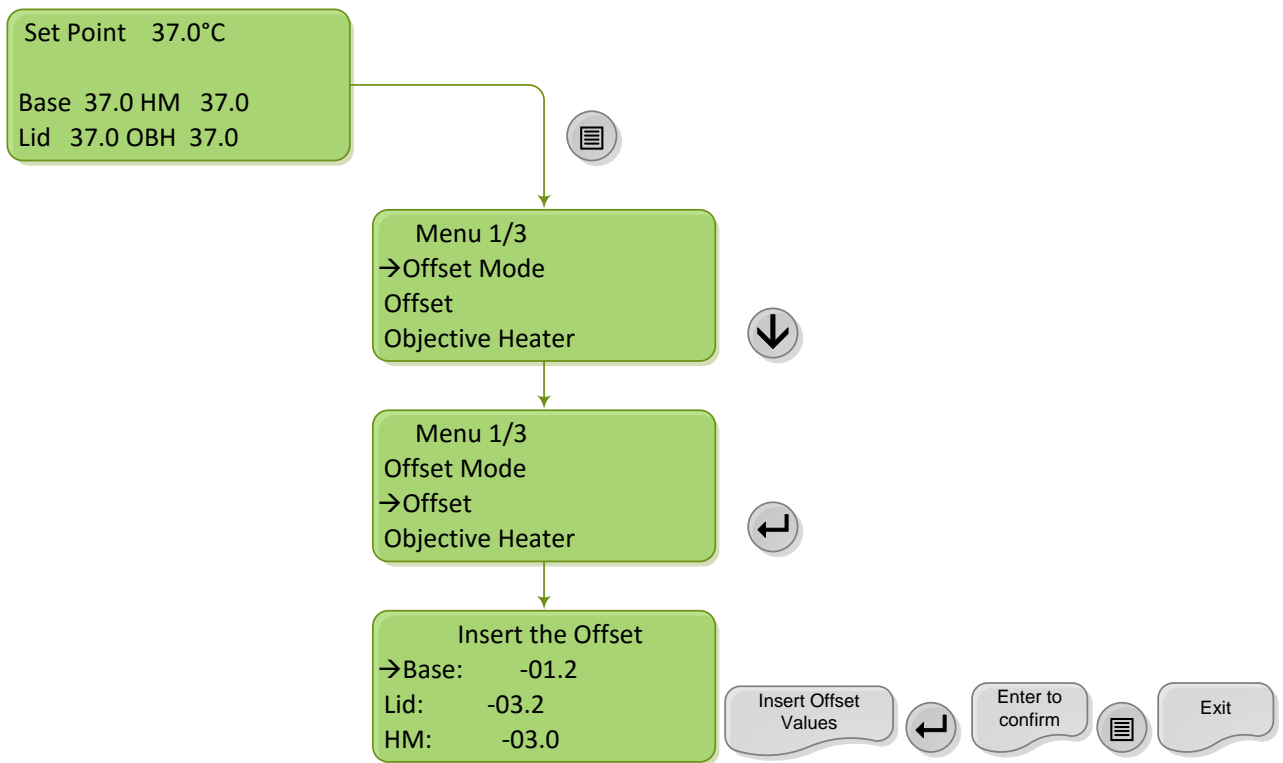


Figure 26. How to insert the custom offset values.

13.6 Objective Heater Submenu

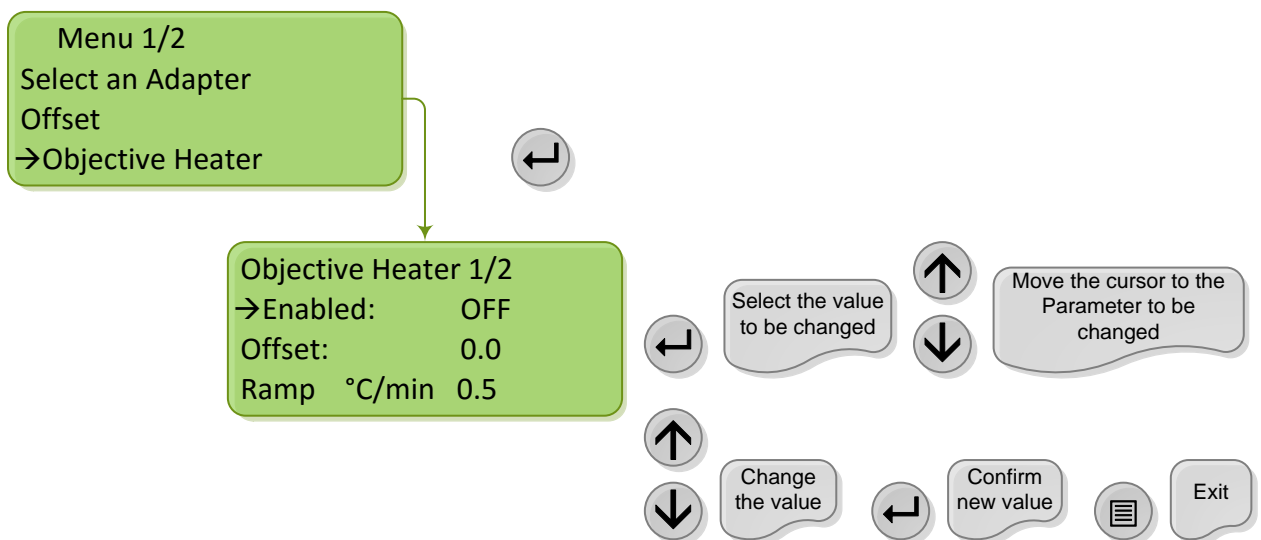


Figure 27. Objective Heater Settings Menu.

- From the main menu press the 'Menu' key on the front panel.
- Move the cursor to the item 'Objective Heater' with the Arrow Keys.
- Press Enter with the cursor on the item 'Objective Heater'.

The User can set the item 'Objective Heater' value to «on» or «off» to enable or disable the Objective Heater. When the Objective Heater is disabled, in the Main Page the Objective

Heater Temperature is no longer displayed, as illustrated in Figure 28 Moreover, in the Objective Heater sub Menu the User can change the offset and select the option 'Reset to factory' to reset Objective Heater Settings to the default values. (See Figure 29)

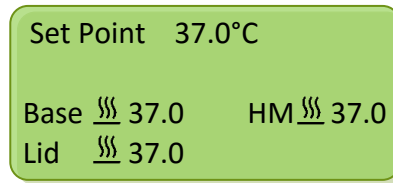


Figure 28. Homepage when objective Heater is disabled.

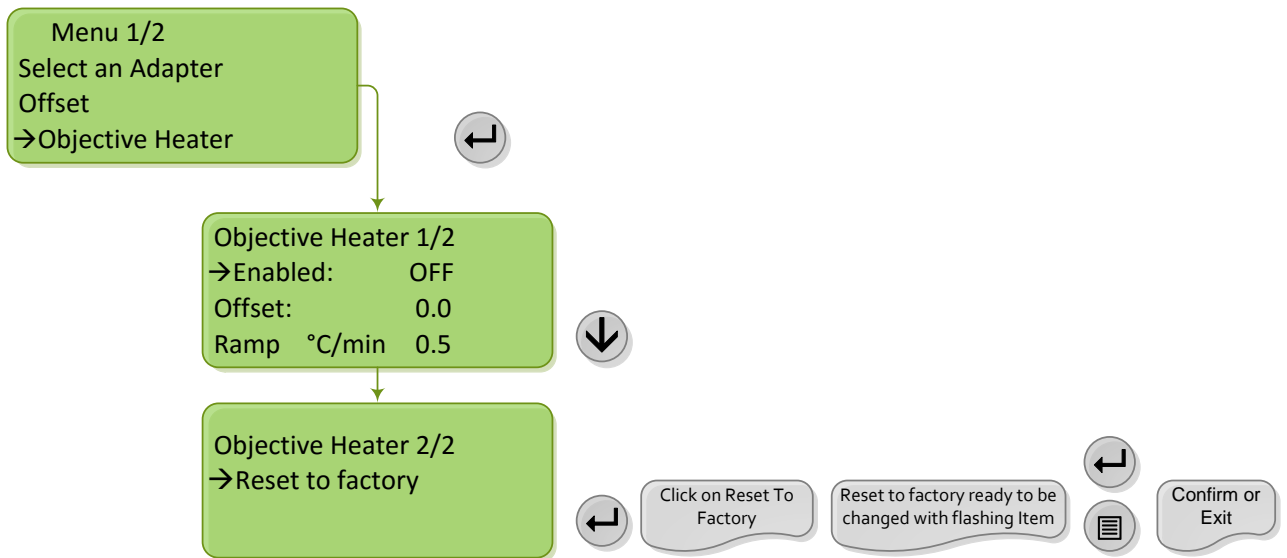


Figure 29. Reset to factory default

In this Submenu, the User can set a manual offset for Objective Heater as described in Figure 30 and following the procedure described in paragraph 14.

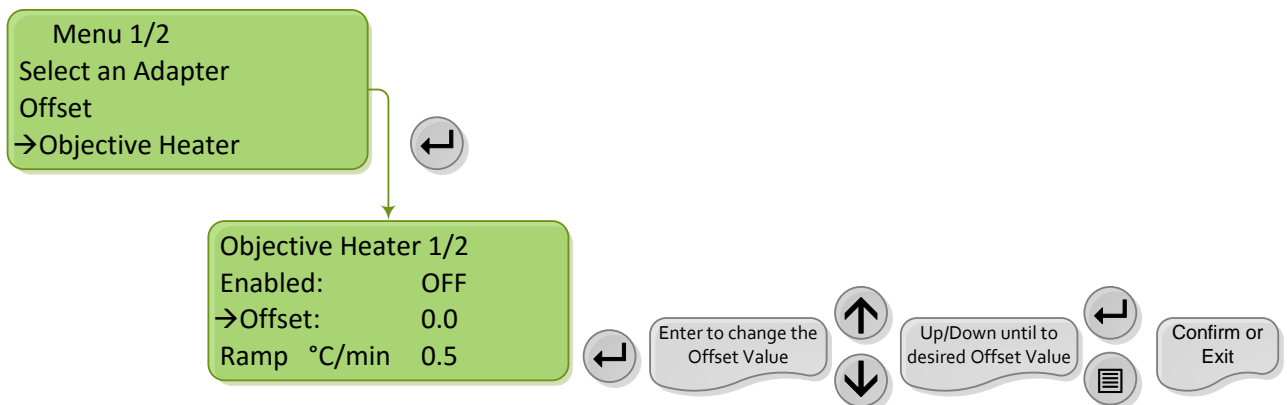


Figure 30. How to change the Objective Heater offset.

- From the main menu press the 'Menu' key on the front panel.
- Move the cursor to 'Objective Heater' using the Arrow Keys.
- Press 'Enter'

- Move the cursor to the item 'Offset' using the Arrow Keys.
- Press 'Enter' with the cursor on the item 'Offset' and then 'Up/Down' to change the offset value
- Press 'Enter' to Confirm

Tip ► The range of the ramp rate values goes from -10°C to 10°C.

The User can change the Ramp rate if needed. This feature allows controlling the heating and cooling rate of the Objective Heater in order to avoid thermal shocks to the sample. To change the Ramp Rate Value see Figure 31.

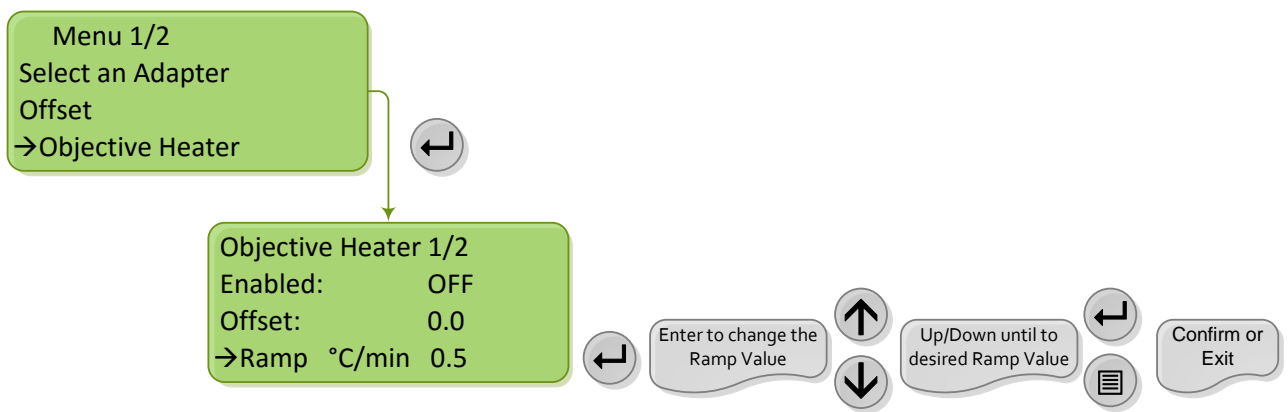


Figure 31. Ramp rate changing.

- From the main menu press the 'Menu' key on the front panel.
- Move the cursor to 'Objective Heater' using the Arrow Keys.
- Press 'Enter'
- Move the cursor to the item 'Ramp' using the Arrow Keys.
- Press 'Enter' with the cursor on the item 'Ramp' and then 'Up/Down' to change the offset value
- Press 'Enter' to confirm

Tip ► The ramp rate can be changed in the range 0.1 °C/min to 1 °C/min.

13.7 Humidity Module Submenu

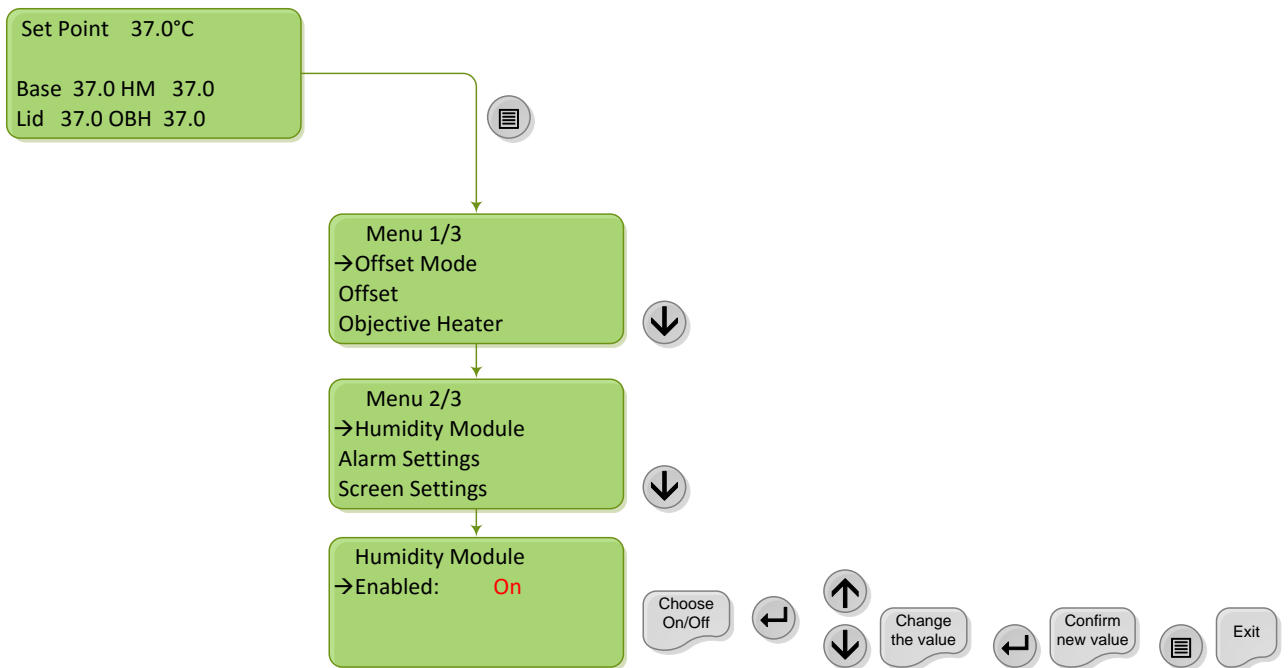


Figure 32. Humidity Module Enabling.

- From the main menu press the 'Menu' key on the front panel.
- Move the cursor to 'Humidity Module' using the Arrow Keys.
- Press 'Enter'
- Press 'Enter' with the cursor on 'Enabled' and then press the Up/Down Arrow keys until you see 'On'
- Press 'Enter' to confirm
- Press the 'Menu' key TWICE to return to Main page.

When the Humidity Module is disabled the main page display will look as in Figure 36.

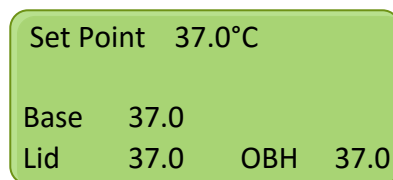


Figure 33. Main Page when Humidity Module is disabled.

13.8 Alarm Settings Submenu

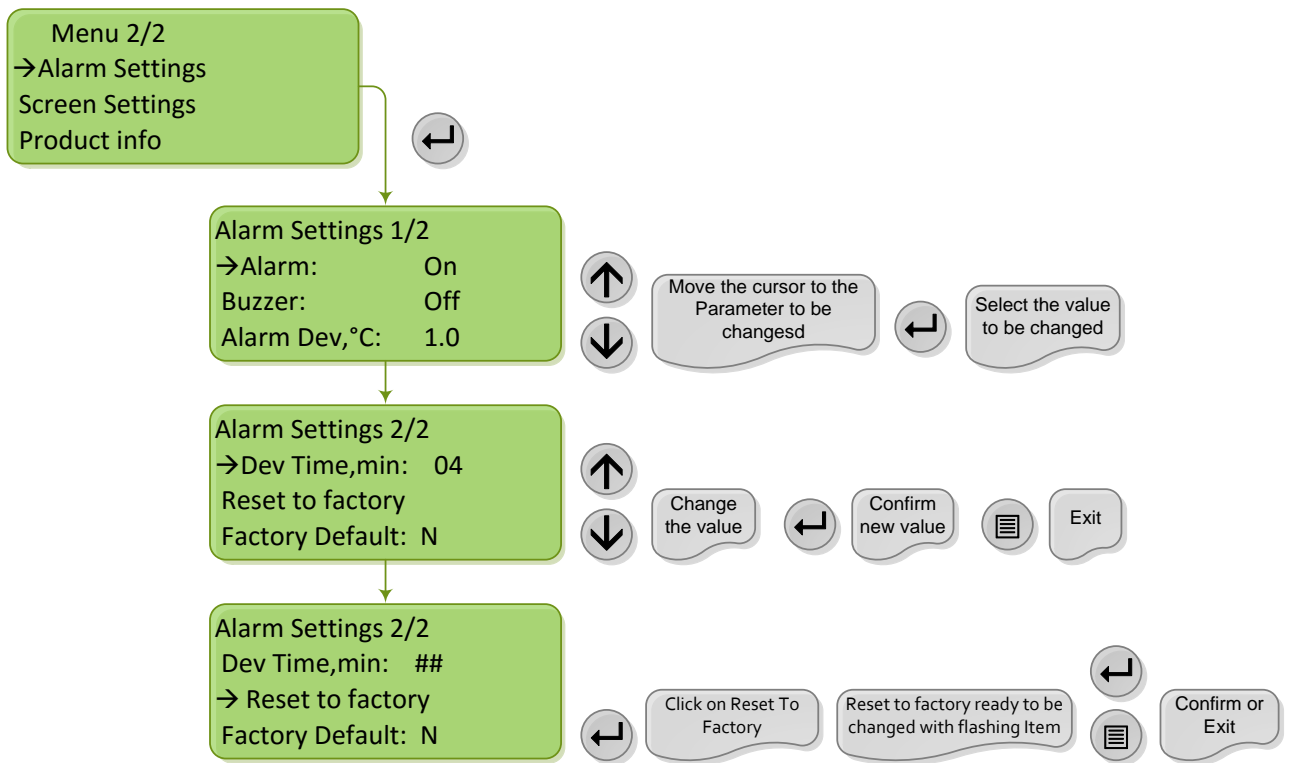


Figure 34. Alarm Settings Menu.

An 'Alarm' can be enabled ('on') or disabled ('off'). With the 'Alarm' 'on' a 'Buzzer' can also be enabled. Right after turning the device on or right after changing the set point temperature, the 'Alarm' will be in standby for 30 minutes, allowing the incubator to reach stable conditions. After 30 minutes the 'Alarm' will activate and start operating. The Alarm function will check for alarm conditions. An exclamation mark (!) will appear next to the component of the chamber with temperature different from the set point more than the **Alarm Dev** and for a length of time longer than **Dev. Time**. (See Figure 35).

Set Point 37.0°C			
Base!	34.0	HM	37.0
Lid	37.0	OBH	37.0

Figure 35. Visual alarm Active.

If the '**Buzzer**' is enabled the sound alarm will go off as well. When 'Alarm' is active and 'Buzzer' is on, an audible alarm is active. Press the 'Up' key one time to stop the buzzer alarm only, for 30 seconds only. If the alarm condition persists, after 30 seconds, the buzzer will go off again.

Set Point 37.0°C			
Base	Err_2	HM	37.0
Lid	37.0	OBH	37.0

Figure 36. Temperature sensor fault.

In Figure 39 above, ERR_2 means that there is no temperature reading from the base of the chamber. This happens if the sensor embedded into the base plate of the chamber is faulty or if its cable is disconnected from the outlet labeled T1. The same ERR_2 appears if Lid or Humidity Module sensor is faulty or disconnected.

13.9 Screen Settings Submenu

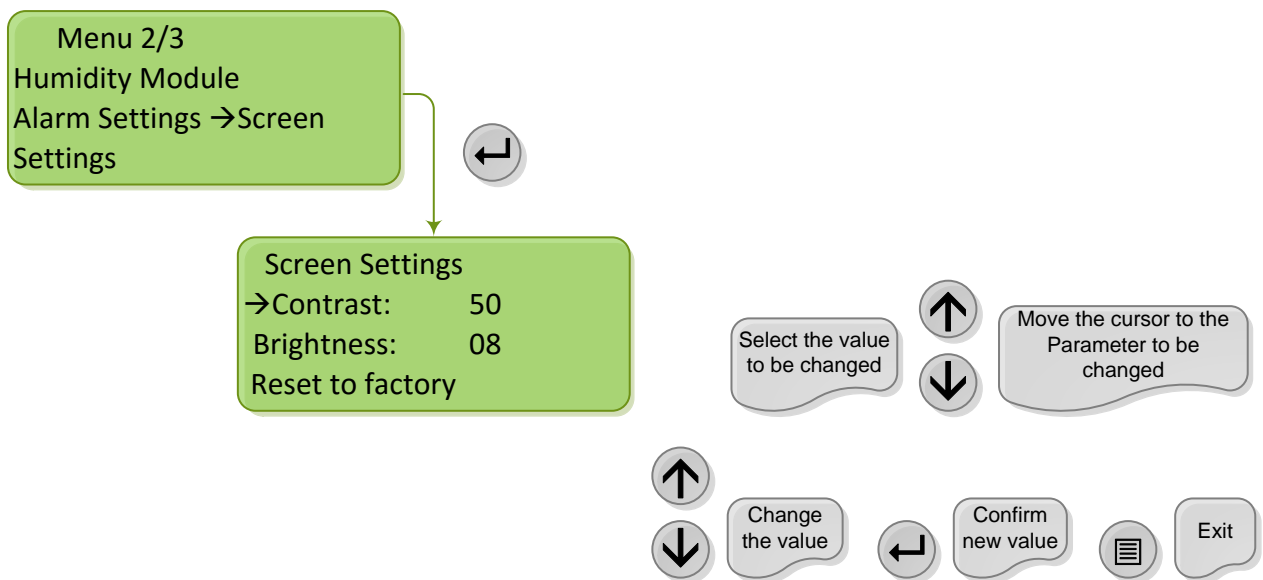


Figure 37. Screen Settings Menu

The Screen Settings Sub Menu enables the operator to adjust the instrument screen contrast and brightness. Use the option 'Reset to factory' to set Screen Settings back to the default values.

- From the main menu press the Menu key on the front panel.
- Move the cursor to 'Screen Settings' using the Arrow Keys.
- Press 'Enter'.

13.10 Product Information Submenu

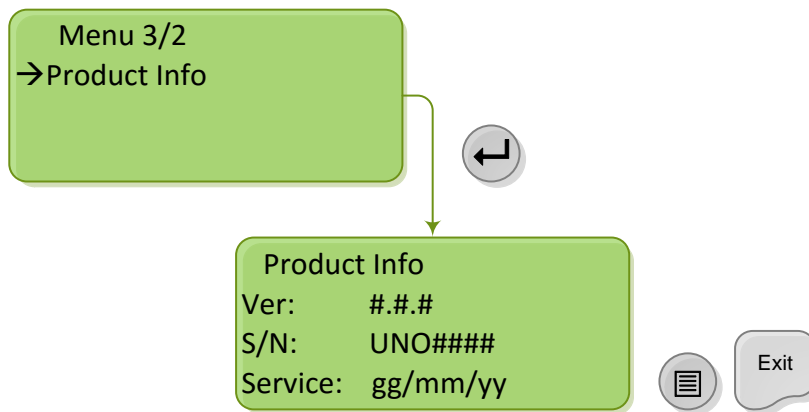


Figure 38. Product Info Menu

Product Info Sub Menu enables the operator to display information such as Software Version (Ver), Serial number (S/N), Service date of the last Factory calibration and set up of the controller.

- From the main menu press the Menu key on the front panel.
- Move the cursor to 'Product Info' using the Arrow Keys.
- Press 'Enter'.

13.11 Stop Mode

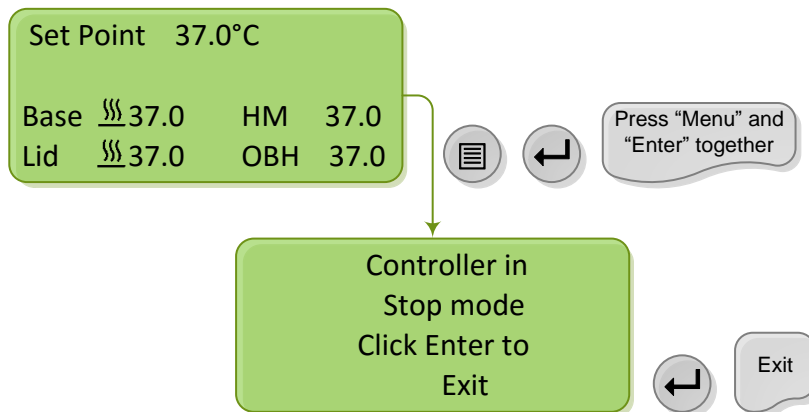


Figure 39. Stop Mode display.

In the Run Mode, press 'Menu' and 'Enter' at the same time to enable Standby Mode. The controller becomes inactive. In Stop Mode the temperature is not controlled and the chamber is no longer heated.

13.12 UNO-TS software Active Message

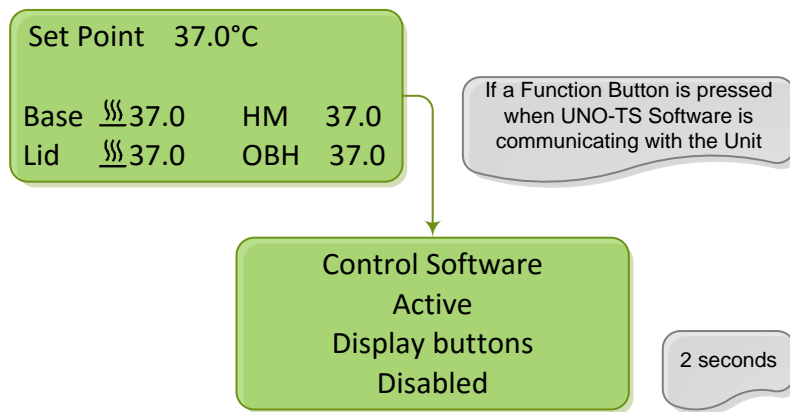


Figure 40. Display when UNO-TS software is active.



When UNO-TS software is active, Menu navigation is disabled

14 Objective Heater Manual Calibration



Carefully read the instructions given in this paragraph and in paragraph 13.6 before starting the Objective Heater Manual Calibration.

The goal of this procedure is to adjust the controlled Objective Heater temperature, manually changing its offset; while maintaining the sample at the desired temperature (e.g. 37.0 °C) once the oil immersion objective is in contact with the bottom of the well. This can be achieved measuring the sample temperature with the Fine Gauge Thermocouple during steady state and estimating the offset values, as follows:

1. Plug the green Fine Gauge Thermocouple into the port labeled "Reference Temperature" located in the rear panel of the Control Unit, as illustrated in Figure 14.
2. Insert the Fine Gauge Thermocouple into the chamber by threading the wire through one of the screw holes on the chamber riser.
3. Secure the end of the Fine Gauge Thermocouple to the bottom of a Petri dish using some tape making sure to avoid covering the head of the probe (see Figure 16).
4. Fill the Petri dish with distilled water to a level sufficient to fully immerse the Fine Gauge Thermocouple. This is a Reference Well simulating experimental conditions. Place the lid

on the Petri dish, and insert it in the incubation chamber. You can also use SENSOR LID-# to close the dish (see the “**Tip**” in 12).

Note ► Please, monitor the room temperature during the calibration and reduce air drafts and forced convection. If possible divert any draft coming from the air conditioning system from the incubation system.

5. Wait until the system reaches the steady state (temperature indicator status is green) then approach the oil immersion objective to the bottom of the Petri dish. The Fine Gauge Thermocouple will detect a decrease in temperature because of the contact with the cold lens. You can see the Fine Gauge Thermocouple temperature decreasing in the Main page (see paragraph Main Page and Main Menu Navigation13.3)
6. When the Fine Gauge Thermocouple reads a stable temperature open the ‘Objective Heater Submenu’ as shown in Figure 27 and select the ‘Offset’ item (See Figure 30) . Next click on “+” or “-” on the ‘Offset’ to adjust the Objective Heater offset value. Okolab recommends starting with an offset value corresponding to the difference between the set point temperature and the actual temperature measured by the Fine Gauge Thermocouple. Press ‘Enter’ and wait until the temperature measured by the Fine Gauge Thermocouple is stable. If the temperature measured by the Fine Gauge Thermocouple is now close to the set point, then the Manual Calibration is complete. If the temperature measured by the Fine Gauge Thermocouple is far from the set point, repeat the steps above until the temperature measured by the Fine Gauge Thermocouple reaches the set point temperature.

15 Flow meter regulation



Do not use the flow meters as closing-opening valves. Once regulated the flow rate (at P=1bar), use the flow meters carefully only for fine regulations

Open the gas valves about 15 minutes before inserting the specimen in the Micro Environmental Chamber. Please, follow carefully scheme in Table 4

Target Gas Percentage [%]		Gas flow rate values to set [NI/min]		
Air	CO ₂	Air	CO ₂	Total
95.00	5.00	0.60	0.03	0.63
95.00	5.00	0.80	0.04	0.84
95.00	5.00	1.00	0.05	1.05

Table 4. Flow meter regulation. NI [Normal/liter].

Tip ► If you need, ask Okolab for personalized Gas Percentage Tables.

'Target Gas Percentage' indicates the stream gas percentage that the operator wants during the experiment.

'Gas flow rate values to set', in Normal liter per minute [NI/min], are the values of gas flow rate to set on the graduated scale (see Figure 41).

'Total' indicates the measured total gas stream flow rate.

Example ► The target Air percentage is 95.00%, the target CO₂ percentage is 5.00%. Table 4, shows three possibilities that reach the required gas percentage. Notice that the values are a compromise between CO₂ consumption, condensation (normally it decreases if Total gas stream flow rate increases), and medium evaporation (normally it decreases if Total gas stream flow rate decreases). In the example below (Figure 44) the gas flows on the second line in Table 4 are shown. Set the flow meters keeping in mind that the correct position of the indicator (floating ball) is about in its middle. The gas stream mix contains, now, 95.00% of Air and 5.00% of CO₂. ◀

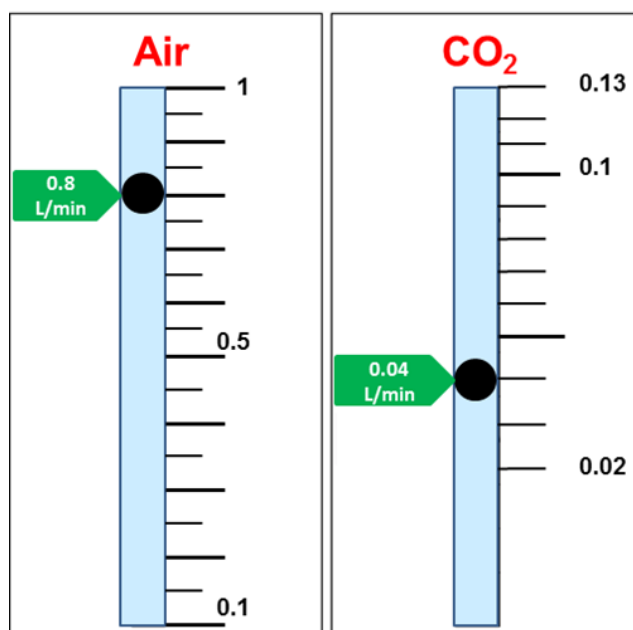


Figure 41. Flow meters graduated scales, example for two gas lines.

If you use a pre-mixed 5% CO₂ gas source connect it to the Air port labeled "Air In" on rear panel of the Control Unit and set the flow rate at the suggested value of 0.3 L/min (See Figure 42).

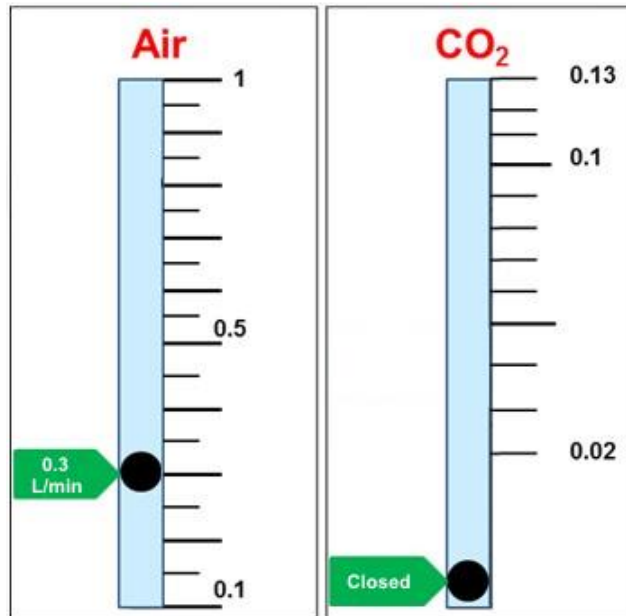


Figure 42. Flow meters graduated scales, example for Premixed as line.

16 Maintenance

To maintain proper Control Unit operation over time:

- Use a polishing cloth or dry cloth to wipe off dust and dirt.



Before cleaning the unit, disconnect the unit from the power source.

Keep away from water.

- Never use thinners, benzene, solvents on or near the devices, since these could corrode their surfaces.
- To clean the Stage Incubator and the Humidifying Module, if it is present, you can use distilled water or alcohol
- Verify the status of all cables and if some cable is damaged, contact Okolab to receive assistance

17 Support

17.1 Web Conference For Assistance And Training

Together with your equipment you received a web cam and headset. You can request remote support over the web. Please follow these guidelines to facilitate setting up your web support:

- Webcam installation (instruction and Cdrom included)
- Updated Skype® software installed (www.skype.com)
- Register yourself on www.skype.com to have an account (Skype_ID)
- Set the audio and video and test them using Skype.
- Contact our technical support (sibillo@oko-lab.com) by e-mail to set an appointment for web assistance.

17.2 Troubleshooting

Incorrect operation of the unit is often mistaken for malfunction. If you think that there is something wrong with a component, see the troubleshooting scheme below. If the problem persists even after troubleshooting as described below, please contact for Okolab support.

Symptom	Probable cause	Remedy
Device off	Supply cable disconnected	Properly connect the cable
	Supply cable damaged	Substitute the cable
	Blown fuse	Replace the fuse (see 'Technical Specifications' paragraph 17.4)
No temperature displayed	Err_1 Cable broken	Contact Okolab to receive assistance
	Err_2 Cable disconnected or free sensor cable damaged	Properly connect the cable or contact Okolab to receive assistance
	Err_3 Error on Free Sensor measure	Check the Thermocouple position Contact Okolab to receive assistance
	Err_4 Temperature read by Thermal plate sensor too high or too low	Contact Okolab to receive assistance
	Err_5 Board Error	Contact Okolab to receive assistance
	Err_6 Generic error	Contact Okolab to receive assistance
Sample temperature differs from the set point	Ambient temperature too low or too high	Regulate the ambient temperature
		Change the Offset value manually
Chamber Base, Lid, Objective Heater or Humidifying Module Heater cold	Low set point value	Change the set point
	Cable disconnected	Connect the cable
	Cable damaged	Contact Okolab to receive assistance
	Heating Elements damaged	Contact Okolab to receive assistance
Alarm rings	Actual temperature far from the Set Point temperature	Quickly shoot down the control unit and properly set the system.
	Cable disconnected	Connect the cable
	Heating elements damaged	Contact Okolab to receive assistance
I check the previous troubleshooting but I cannot solve the problem		Contact Okolab to receive assistance

17.3 Technical Support

Please, do not hesitate to contact Okolab should you need any further commercial information or technical support.

Please, check Okolab web site www.oko-lab.com for news, events, new products and general FAQ.

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Okolab S.r.l.

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17.4 Italy Technical Specifications

UNO –COMBINED.CONTROLLER – Technical Specifications	
Temperature	Chamber Temperature Range: 25-50°C Sample Temperature range: 3°C above ambient temperature to 50°C Step size: 0.1°C Accuracy: 0.3°C if room temperature remains within $\pm 1^\circ\text{C}$
Regime temperature time	about 30 min
Operating Temperature	0°C ~ +55°C
Storage Temperature	-5°C ~ +60°C
Operating Humidity	0-70%
Power Consumption	115V AC 60 Hz Fuse 2.5 AT 250V 5x20 230V AC 50 Hz Fuse 1.25 AT 250 V 5x20 160 W max
Output	Fuse 1 24 VOC 2.5 A Max Current Fuse 2.5 AF 250V 5x20 Fuse 2 24 VOC 1.6 A max Current Fuse 1.6 AF 250V 5x20 Fuse 3 24 VOC 1.6 A Max Current Fuse 1.6 AF 250V 5x20 Fuse 4 24 VOC 1.6 A Max Current Fuse 1.6 AF 250V 5x20
Weight	5 Kg (only the Control Unit)

18 Figures

Figure 1 AC section wiring diagram	10
Figure 2. System Overview.	11
Figure 3. Control Unit Rear Panel Overview.....	15
Figure 4. Complete gas lines scheme.	16
Figure 5. Complete gas lines scheme. Zoom sketches on the Inlet and Outlet connectors of the Humidifier and Zoom sketch on the Inlet connector of the Chamber are reported.	17
Figure 6. Avoid gas leak: make sure tubing is pushed all the way into the Push-in to-Fit connector.....	18
Figure 7. Input and Output gas ports. See symbol on the rear of Pressure Gauge.	19
Figure 8. How to close the purging valve of the Pressure Gauge.	20
Figure 9. Pressure gauge using.	20
Figure 10. Assembling instruction of the Bubbling Column and its parts	21
Figure 11. Cables connection. Standard configuration.....	22
Figure 12. Objective Band Heater Selection Chart.	23
Figure 13. Objective Heater (OBJ-COLLAR ####) Assembly	24
Figure 14. Free Sensor with male to male extending cable.	27
Figure 15. Front Panel of the Control Unit	28
Figure 16. Free Sensor attached on the internal base of the Petri Dish.....	31
Figure 17. SENSOR LID-35. For 35 mm Petri-dish	32
Figure 18. Controller Display "Main Page" and function buttons	34
Figure 19. Main Page and Main Menu Navigation Description	36
Figure 20. Main Menu Display. Changing Set Points Description.....	37
Figure 21. Main Page when working with Free Sensor connected to the Control Unit.	37
Figure 22. Offset Mode Selecting.....	38
Figure 23. How to see the current offsets in standard mode.....	38
Figure 24. How to change the base offset value.	39
Figure 25. How to choose the Custom Offset Mode	40
Figure 26. How to insert the custom offset values.	41
Figure 27. Objective Heater Settings Menu.....	41
Figure 28. Homepage when objective Heater is disabled.	42
Figure 29. Reset to factory default	42
Figure 30. How to change the Objective Heater offset.....	42
Figure 31. Ramp rate changing.	43
Figure 32. Humidity Module Enabling.....	44
Figure 33. Main Page when Humidity Module is disabled.	44
Figure 34. Alarm Settings Menu.	45
Figure 35. Visual alarm Active.	45
Figure 36. Temperature sensor fault.	46
Figure 37. Screen Settings Menu	46
Figure 38. Product Info Menu	47
Figure 39. Stop Mode display.	47
Figure 40. Display when UNO-TS software is active.	48
Figure 41. Flow meters graduated scales, example for two gas lines.	50
Figure 42. Flow meters graduated scales, example for Premixed as line.....	51

WARRANTY

Okolab S.r.l. warrants its products to be free of defects in materials and workmanship for a period of one year starting from invoice date. If the unit malfunctions, it must be returned to the factory for evaluation. If the equipment has to be returned to the factory, please ensure that is carefully and properly packed. Okolab S.r.l. accepts no responsibility for damage due to unsatisfactory packing.

Upon examination of Okolab S.r.l., if the unit is found to be defective, it will be repaired or replaced at no charge. This warranty does not apply to defects resulting from any actions of the purchaser. Okolab S.r.l. neither assumes responsibility for any omissions or errors nor assumes liability for any damages that result from operation not according to the instructions listed in the operation manual. This warranty does not cover or involve any other equipment that may be used along with the Okolab System (i.e. mini-incubators, any gas tank, etc.), whose usage should be considered independent and performed according to their own operational instructions.

Okolab S.r.l. warrants only the parts manufactured by it will as specified and free of defects. Okolab S.r.l. makes no other warranties or representations of any kind whatsoever, express or implied, except that of title, and all implied warranties including any warranty of merchantability and fitness for a particular purpose are hereby disclaimed. LIMITATION OF LIABILITY: the total liability of Okolab S.r.l. shall not exceed the purchase price of the component upon which liability is based. In NO event shall Okolab S.r.l. be liable for consequential, incidental or special damages.