

LEO

Handheld Analyzer for IVF Applications

Manual

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1 Symbol description
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This paragraph describes the symbols used in this manual and on the product label.

1.1 Symbols used in this manual and on the product

The following symbols identify important information:



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

Tip ► *Supplies you with helpful suggestions.*

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

1.2 Symbols on the product label



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



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



This symbol indicates the product production date.



This symbol indicates the Manufacturer data.



This symbol warns you to read the user manual before starting the device.



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

2 Safety Notes

In order to achieve maximum performance and to ensure proper operation of your new equipment, please read carefully the following safety notes and the instructions. If you have any question, please contact Okolab.

- This equipment must be used only as intended and described in this Manual.
- This equipment should be operated only by qualified personnel.
- Do not use this equipment if any parts are damaged.
- This instrument is not intended for use in locations subject to flammable or explosive gases.
- Always transport equipment with care.
- The equipment and its internal parts may be damaged by dropping or 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 could damage the device and its accessories.
- Do not exceed the voltages indicated in this manual or on the product label.
- Avoid excessive induction noise, static electricity, or magnetic fields.
- Do not expose this instrument to water or moisture.
- Avoid throttling and kinking of tubes.
- Check tubing from time to time for possible wear and tear.
- Check that all tubing is fully inserted into connectors, and secure.
- This device is not designed for use in medical applications.



International caution symbol marks this device. It is important to 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 upgrades and changes.

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



3 Regulatory Compliance

3.1 Regulatory Statement

The product complies with the essential requirements of the LVD Directive 2014/35/EU and the EMC Directive 2014/30/EU. The product has been tested and it complies with the requirements of the following standards:

- European standard EN 61010-1: 2010 "Safety requirements for electrical equipment for measurement, control and laboratory use"
- European standard EN 61326-1: 2013 "Electrical equipment for measurement, control and laboratory use – EMC requirements"

The product meets the technical requirement of the Directive 2011/65/EU Restriction of the use of certain hazardous substances (RoHS). The product has been tested according to European Standard EN 50581: 2012 "Technical documentation for the assessment of the electrical and electronic products with respect to the restriction of hazardous substances".

3.2 Waste Electrical and Electronic Equipment Directive (WEEE Directive)

The product must not be disposed as a general household waste. At the end of the product's life, take the product to a collection point designed for recovering and recycling of electrical and electronic devices.

In case of doubt, please return the product to Okolab s.r.l. for proper treatment.

4 Preface

LEO (see Figure 1 for front and rear view of LEO) is a handheld meter for IVF Applications, allowing measurement of CO2-O2 gas composition, temperature, relative humidity, gas flow of the high-top and bench-top incubators, by using multiple sensor modules.

The CO2-O2-MODULE is embedded into LEO, and it is included standard. It measures, with a high accuracy, the CO2-O2 volume concentration in the unit of %, and CO2-O2 partial pressure in the units of mbar, psi and KPa.

Other (external) sensor modules that can be connected to LEO's connection ports are: T1-MODULE, T2-MODULE, CO2-PPM-MODULE, HUMIDITY-MODULE, FLOW-RATE-MODULE. All sensor modules are calibrated by using traceable standards, and store the calibration data in the internal memory. The sensor modules can be delivered with traceable or accredited calibration certificate, and sent to Okolab for re-calibration. Moreover, sensor calibration can be performed by using LEO intuitive user calibration routines.

LEO works in Diffusion Mode when the gas to be sampled is pressurized and in Aspiration Mode when it works at atmosphere pressure, sucking the gas from the incubator by an internal pump. In both cases, if the gas inside the incubator is humid, prior de-humidification with the Moisture Trap (included standard with LEO) is needed to retain water molecules.

LEO can improve the CO2 and O2 reading accounting the water content of your incubator by use Wet Mode. The temperature and relative humidity (RH) values of your incubator must be entered.

LEO can perform both single measurement and extended time logging, storing the data in its local memory. LEO allows organization of the stored data, associating a label to each device. You can store up to 40 devices labels. The stored data to be downloaded through the micro USB port, and analyzed by using the free LEO DATA IMPORT Excel Macro.

LEO features a user-friendly 4.3" touch screen interface.





Figure 1. LEO: front (a) and rear (b) view.

4.1 Sensor Modules

The available Sensor Modules (see paragraph 10 and paragraph 13.4)

- CO2-O2-MODULE (embedded into LEO)
- T1-MODULE (external)
- T2-MODULE (external)
- CO2-PPM-MODULE (external)
- HUMIDITY-MODULE (external)
- FLOW-RATE-MODULE (external)



Figure 2. LEO with T1-MODULE and T2-MODULE

5 Supplied equipment

Figure 3 shows the Hard Case containing LEO's equipment. The optional and spare parts can be purchased from Okolab s.r.l. Please see <u>www.oko-lab.com</u> or contact <u>info@oko-lab.com</u> for more details.



Figure 3. LEO Hard Case.

- A **LEO**. Hand-held meter for IVF applications.
- B **Power Supply** (see paragraph 5.1).
- C **USB Cable**.
- D **Soda Lime Kit** (see paragraph 5.2).
- E **Moisture Trap**, for humid gas measurement (see paragraph 5.3).
- F **Filter**
- G **OTG Cable** (see paragraph 5.4).
- H Lanyard.
- I **Fitting Tubes** (see paragraph 5.5).
- J **Tube-B** (see paragraph 5.8).
- K **Tube-A** (see paragraph 5.7).

- L **Calibration Kit** (see paragraph 5.6).
- M Additional tubes

5.1 Power Supply

LEO is powered by a rechargeable battery. A power supply is included with the device for charging the battery from a power outlet. Connect the USB cable between the LEO MICRO-USB port and the power adapter to charge the internal battery in LEO (see Figure 4).



Figure 4. LEO connection for battery recharging.



Figure 5. Power Adapters supplied.



Do not use a different power supply to recharge LEO. Do not attach a different USB cable to the power adapter.

5.2 Soda Lime Kit

The Soda Lime Kit (D in Figure 3) is used for the *CO2 Zero Reset* procedure with ambient air (see paragraph 13.4.1). It consists of a Soda Lime filter (1 in Figure 6) generally used to remove CO2 from the ambient air.

The Soda Lime Kit must be connected to Tube-A to start the *CO2 Zero Reset* procedure using ambient air. The **clear connector** (2 in Figure 6) of Soda Lime Kit has to be connected to the **red connector** of Tube-A (1 in Figure 10).

Note ► The Soda Lime kit has to be replaced when the Soda Lime filter (1 in Figure 6) changes color turning purple. Contact Okolab to purchase a new Soda Lime Kit.

Note ► *Remove the protection cap from the Soda Lime filter before using the Soda Lime Kit.*



Figure 6. Soda Lime Kit.

5.3 Moisture Trap

The Moisture Trap (E in Figure 3) is used for humid gas measurements. Figure 7 shows the Moisture Trap components:

- A condensation trap allowing for the reduction of sampled gas temperature and the collection of condensed water from the sampled gas (4 in Figure 7). The condensation trap top (3 in Figure 7) allowing reducing the sampling gas time during the Single Point measurement must be removed when Gas Logging occurs.
- 2. A Selemion Tube that reduces the relative humidity of sampled gases to a value lower than 40% (5 in Figure 7).

The **connector marked in green** is the Gas Inlet port of the Moisture Trap (1 in Figure 7). Please use Tube-B to connect the sampling Gas Port of the incubator to Gas Inlet port of the Moisture Trap.

The **connector marked in red** is the Gas Outlet port for the Moisture Trap (2 in Figure 7). Please use the Tube-A to connect the Gas Outlet port of the Moisture Trap to the Gas Inlet port of LEO (see Configuration b in Figure 46 for the measurements in Diffusion Mode or Configuration b in Figure 48 for the measurements in Aspiration Mode).



Figure 7. Moisture Trap.

5.4 OTG-Cable

The OTG Cable (G in Figure 3) can be used to download data stored on LEO to a USB Key. USB Key is not supplied.



Figure 8. OTG Cable

5.5 Fitting Tubes

Okolab supplies 5 different types of reduction fitting tubes (I in Figure 3) allowing the compatibility of its tubes with many sampling gas ports of the incubators.

Okolab provides the following reduction fitting tubes:

- 1. from ID=1.6mm to ID=3.2mm
 - 2. from ID=2.4mm to ID=3.2mm
 - 3. from ID=4.0mm to ID=3.2mm
 - 4. from ID=4.8mm to ID=3.2mm
 - 5. from ID=6.4mm to ID=3.2mm

Three additional caps for LEO's ports, a male and female luer connector have been also supplied.

Note ► Additional caps are used to perform the Leak test of the pump (see paragraph 9.3.3.1).

5.6 Calibration Kit

The Calibration Kit (L in Figure 3) allows calibration of CO2-O2-MODULE.

Figure 9 shows the Calibration Kit. The **clear connector** (1 in Figure 9) has to be connected to **red connector** of TUBE-A (1 in Figure 10). The end of Calibration Kit (3 in Figure 9) has to be connected to the gas

cylinder. The calibrated orifice (2 in Figure 9) guarantees a gas flow rate of 150 ml/min when the output pressure from the gas cylinder is set at 1.0 barg (14.5 psig).

The relief valve (4 in Figure 9) allows avoiding over pressurization.

For more details on the connections and the use of the Calibration Kit, see the paragraph 11.

Note ► *Do not exceed 1.5 barg (22 psig) during the calibration procedure (see paragraph 11).*

Note ► Remove the protection caps from Calibration Kit before using it.



Figure 9. Calibration Kit

5.7 Tube-A

Tube-A (K in Figure 3) is used to connect the Gas Inlet connector of LEO to the Tube-B if the gas to be sampled is dry or to the Gas Outlet connector of the Moisture Trap, if the gas is humid.

Figure 10 shows Tube-A. Connect the **red connector** (1 in Figure 10) to the green end of Tube-B (1 in Figure 11) when sampling a dry gas or to the Gas Outlet connector (labeled green) of the Moisture Trap (1 Figure 7) when sampling a humid gas. The **clear connector** of Tube-A (3 in Figure 10) must be connected to one of the Gas Inlet connector of LEO (see Figure 46 for the measurement in Diffusion Mode or Figure 48 for the measurements in Aspiration mode). A hydrophobic filter (2 in Figure 10) is fitted in order to avoid water collection at the Gas Inlet.





Note ► Okolab suggests to replace the filter every six months.

Note ► *Remove the protection caps from Tube-A before using it.*

5.8 Tube-B

Tube-B (J in Figure 3) is used to connect the sampling Gas Port to the Tube-A if the sample gas is dry, or to the Gas Inlet connector of the Moisture Trap if the sample gas is humid.

Figure 11 shows Tube-B. The **green connector** (1 in Figure 11) must be connected to the **red connector** of TUBE-A (1 in Figure 10) when sampling the dry gas or to the Gas Inlet (labeled green) connector of the Moisture

Trap (1 in Figure 7). The end of Tube-B (2 in Figure 11) must be connected to the sampling Gas Port of the incubator.





Note ► Remove the protection caps from Tube-B before using it.

5.9 Additional tubes

Okolab supplies the following additional tubes (M in Figure 3):

- 1. Tygon tube (OD = 6mm) with a length of 100 cm
- 2. Tygon tube (OD = 4 mm) with a length of 30 cm
- 3. Silicone tube (OD = 4mm) with a length of 10 cm

5.10 CO2-O2-MODULE (embedded in LEO)

The CO2-O2-MODULE is embedded into LEO, and it is included standard. It measures, with a high accuracy, the CO2-O2 volume concentration in the unit of %, and CO2-O2 partial pressure in the units of mbar, psi and KPa. CO2-O2-MODULE is temperature and pressure compensated thanks to a dedicated temperature and pressure sensor that are included in this module.

The CO2-O2-MODULE is factory calibrated by using traceable standards, and stores the calibration data in its internal memory. The declared accuracy is guaranteed for a period of 6 months from the factory calibration date. The CO2-O2-MODULE is designed for an easy extraction from LEO (see paragraph 14).

The CO2-O2-MODULE lifetime is 5 years.



Figure 12. CO2-O2-MODULE

5.11 Pump (embedded in LEO)

The pump is a micro diaphragm pump that guarantee the desired gas flow rate (from 60 ml/min to 260 ml/min). The default flow rate is 150 ml/min. The pump is designed for easy extraction from LEO, see paragraph 18). Figure 13 shows an image of the pump inside LEO.



Figure 13. Pump

6 Optional Parts

- 1. T1-MODULE
- 2. T2-MODULE

6.1 T1-MODULE

The T1-Module (see Figure 14) is a calibrated temperature probe that can be read and displayed by LEO. The T1-MODULE has an external diameter of 0.13 mm and it's suitable for temperature measurements of benchtop and hightop incubators where the temperature monitoring port has a diameter smaller than 2mm. T1-MODULE stores calibration data in its own local memory

Tip \blacktriangleright Calibration data is available in the Calibration page of LEO when T1-MODULE is connected to LEO (see paragraph 12).



Figure 14. T1-MODULE

Note ► Okolab recommends that the T1-MODULE should be calibrated after one year of use.

6.2 T2-MODULE

The T2-MODULE (see Figure 15) is a calibrated temperature probe that can be read and displayed by LEO. The T2-MODULE has an external diameter of 2 mm and it's suitable for temperature measurements of benchtop and hightop incubators where the temperature monitoring port has a diameter bigger than 2 mm. T2-MODULE stores calibration data in its own local memory.

Tip \blacktriangleright Calibration data is available in the Calibration page of LEO when T2-MODULE is connected to LEO (see paragraph 12).



Figure 15. T2-MODULE

Note ► Okolab recommends that the T2-MODULE should be calibrated after one year of use.

All connection ports of LEO are illustrated in Figure 16.



Figure 16. LEO connections

LEO features:

1. Power button

There are many different press modes:

- To turn LEO on: press and hold the power button for 2 seconds.
- To turn LEO off: press and hold for 2 seconds the power button, and tap *OK* to confirm.
- Press and hold for 5 seconds the power button to start the Touchscreen Calibration procedure (see paragraph 15).
- Press and hold for 10 seconds the power button to restart LEO.
- Press the power button to lock and unlock LEO's screen during the use.
- Press the power button to display the battery charging status (see paragraph 8.1).
 - 2. MICRO-USB port. To charge the battery with the power adapter (see paragraph 5.1) and to download software updates
 - 3. Gas input port labeled with icon =
 - 4. Gas input port labeled with icon IN
 - 5. Probe connector 1. To connect additional external sensor modules
 - 6. Probe connector 2. To connect additional external sensor modules
 - 7. Gas outlet for gas release during the gas sampling measurement

8 Preliminary operations

8.1 Charge the battery

To charge the battery, connect the power adapter to the MICRO-USB port (2 in Figure 16). If the battery is being charged while LEO is turned on, the *Charging* symbol ⁽²⁾ appears at the top of the LEO display (1 in Figure 17). The percentage of battery charge shows besides the battery symbol (2 in Figure 17). When charging is complete, the *Battery full* symbol ⁽²⁾ appears at the top of display.



Figure 17. Homepage - battery

If LEO is switched off, press the power button to display the battery charging status and the percentage of battery charge (see Figure 18).



Figure 18. Battery charging status.

Note \blacktriangleright If you charge LEO with computer, the screen in Figure 18 a appears that means that LEO is not getting the proper charging power that it expects.

Note ► A full charge requires 4 hours. The recommended initial charging time is 6 hours.



Okolab recommends not to use LEO during the first charging.

Note \blacktriangleright Okolab suggests to use LEO while charging the battery only if the percentage of battery charge is higher than 90%.

8.2 Date & Time setting

When turning LEO on for the first time, Date & Time have to be set because the current date & time values are stored with every reading. To change Date & Time and their format see paragraph 9.3.5.1.

8.3 Memory Check

When you want to use LEO for a large number of measures, Okolab recommends checking the system memory. LEO has a memory page where the stored records and the still available records are shown (see paragraph 9.3.5.3).

In the System Memory 300 records can be stored. The memory page shows how many records can be saved. To check memory status, follow the indications described in the paragraph 9.3.5.3.

9 Navigation menu

9.1 Homepage

Figure 19 shows the Homepage of LEO:



Figure 19. LEO Homepage

- 1. **Wet/Dry button**: to set the Dry or Wet Mode. See paragraph 10.1.5.
 - 2. Pump/Diffusion button. Press Pump button to run the pump and draw the gas sampling into LEO
 - 3. **CO2-O2 Single Point Measurement**: to start a Single Point measurement of CO2 -O2 gas concentration. See paragraph 10.1.3.
 - 4. **CO2.O2 Logging**: to start a Logging of CO2 -O2 gas concentration. See paragraph 10.1.4.
 - 5. **Temperature Single Point Measurement**: to start a Single Point temperature measurement. See paragraph 11.2. If no Temperature sensor is connected to LEO, this function does not work.
 - 6. **Temperature Logging**: to start a temperature logging. See paragraph 11.3. If no Temperature sensor is connected to LEO, this function does not work.
 - 7. **Device**: to select (or add) a device. See paragraph 9.3.1.
 - 8. **View**: to access to the stored data. See paragraph 12.1.

- 9. Settings: to access LEO's settings. See paragraph 9.3.
- 10. Home: to return to the Homepage.
- 11. T2: T2 measurement. Not displayed if no T2-MODULE is connected to LEO.
- 12. **T1**: T1 measurement. Not displayed if no T1-MODULE is connected to LEO.
- 13. Status of temperature measurement. One of the following five options will be displayed:
 - "NO sensors" (if no sensor is connected)
 - "NO measure" (if no measurement is in progress)
 - "Single point" (if a single point temperature measurement is in progress)
 - "Logging" (if a Temperature Logging is in progress)
 - *"Completed"* (if a Temperature Logging is completed)
- 14. Device name: the name of the device for which data will be stored (see paragraph 9.3.1).
 - 15. **02**: 02 measurement.
 - 16. Alarm Icon: Device alarm. See paragraph 9.2.
 - 17. **CO2**: CO2 measurement.
- 18. Status of CO2-O2 measurement. One of the following five options will be displayed:
 - "NO measure" (if no CO2-O2 measurement is in progress)
 - "Single point" (if a single point gas measurement is in progress)
 - "Logging" (if a CO2-O2 Logging is in progress)
 - "Completed" (if a CO2-O2 measurement Logging is completed)
 - 19. Device name: the name of the device for which data will be stored (see paragraph 9.3.1).
 - 20. PC Icon: PC connection.
 - 21. Alarm Icon: Device alarm. See paragraph 9.2.
 - 22. Pump Icon: Displayed only when the pump is on.
 - 23. Battery Status with percentage of battery charge
 - 24. Time.
- 25. USB Icon: Displayed when a USB key is connected to LEO for data download. See paragraph 12.2.
- 26. RH: Relative humidity measurements. Not displayed if no HUMIDITY-MODULE is connected to LEO
- 27. T: Gas temperature measurement, during if no HUMIDITY-MODULE is connected to LEO.
- 28. **CO2 ppm**: to measure carbon dioxide at ppm. Not displayed if no CO2-PPM-MODULE is connected to LEO.
- 29. VOC: VOC measurement. Not displayed if no VOC-MODULE is connected to LEO.

9.2 Alarm Icons and meanings

LEO can display different alarms, which are identified by the Alarm Icons displayed on the Homepage during an alarm condition.



The Yellow Warning Alarm Icon (15 in Figure 19) indicates that the current CO2 or O2 value is out of the range defined in the alarm subpage (see paragraph 9.3.4) during the Transient Time. The meter doesn't trigger in Alarm and no action is needed. As soon as the CO2 or O2 value is within the defined range, the Yellow Warning Alarm Icon disappears. If after the Transient Time the CO2 or O2 value is still out of the range defined by the operator, the Yellow Warning Alarm Icon changes to Red Warning Alarm Icon.

 $Tip \triangleright$ If the temperature sensors are connected to LEO, the Yellow Warning Alarm Icon appears next to T1 and/or T2 during an alarm condition.



The *Red Warning Alarm Icon* indicates that the current CO2 or O2 value is out of the range defined in the alarm subpage (see paragraph 9.3.4) after the Transient Time. The meter triggers in Alarm.

Tip
ightarrow If the temperature sensors are connected to LEO, the Red Warning Alarm Icon appears next to T1 and/or T2 during an alarm condition.



The *Humidity Alarm Icon* indicates that the current humidity value in LEO is higher than 80% (for example the Moisture Trap is full). The meter triggers an Alarm. The Alarm Icon disappears when the humidity returns to a value lower than 80%. This alarm is always active during all type of measurements. If the alarm occurs during a measurement in Aspiration Mode, stop the Pump. Failing to do so will expose the CO2 O2 sensor to moisture damaging it.

9.3 Settings

Press on *Settings* icon ^(C) (see Figure 20 a) to enter the *Settings* menu, as shown in Figure 20 b. The settings menu has eight items:

- 1. Device. See paragraph 9.3.1
 - 2. Calibration. See paragraph 9.3.2
 - 3. Pump. See paragraph 9.3.3
 - 4. Alarm. See paragraph 9.3.4
 - 5. System. See paragraph 9.3.5
 - 6. Display. See paragraph 9.3.6
 - 7. Units. See paragraph 9.3.7
 - 8. Factory reset. See paragraph 9.3.8



Figure 20: Settings Page (a – b).

9.3.1 Device

LEO allows associating a label to each device connected. To choose an existing device press the Device icon

in Settings page (see Figure 21 a) and select the device-name for which data shall be stored (see Figure 21 b). When selecting an already existing device, a pop-up dialog box appears (see Figure 21 c) showing the three following options:

- *Use*: to use the selected device.
- *Edit*: to change the device name.
- *Delete*: to delete the device.



Figure 21: Device selection (a – b - c).

Note ► *When deleting a device, all its data is erased too.*



You can also add a new device by clicking on "*Add*", as shown in Figure 22 a. Write the device name by using the keyboard that will appear on the display (see Figure 22 b) then press "*Save*". You can add up to 40 devices.

94% 🗐 12:56	71% 🚱 09:39
Device	Device
1 - Default	Add Device
2 - incub 1	
3 - incub 2	
4 - incub 3	
5 - incub 4	asdfghjk1 zxcvbnm < AB 12#
Add	Cancel rotate Save
Home Settings View Device	Home Settings View Device
а	b

Figure 22: How to add a new device (a - b).

9.3.2 Calibration

By pressing the Calibration icon (as shown in Figure 23 a) you can calibrate the available sensors and you can display stored calibration data.



Figure 23. Calibration page (a – b).

Note ► *External sensor modules' icons are automatically disabled if sensor modules are not connected to LEO.*

9.3.3 Pump

Figure 24 shows the internal pump subpages. By pressing *Pump* icon ⁴ (see Figure 24 a), you can read the remaining lifetime of the pump and its current flow rate.

Press Advanced (see Figure 24 b) to set a different flow rate (from 60 to 260 ml/min) (see Figure 24 c).

Tip ► *The default flow rate value is 150 ml/min.*

You can also reset the remaining lifetime if a new pump is installed by pressing on "*Reset*" (see Figure 24 c). See the paragraph 18 for the pump replacement.

Note ► *The pump lifetime must be reset only if you replace the pump with a new one.*



Figure 24. Pump Page (a – b - c).

9.3.3.1 Pump Leak Test

You can perform the Leak test to verify the proper functioning of the pump.



Figure 25. Leak test selection (a - b - c).

After pressing on Leak test, follow the instruction below for running the test.

Tip ► *LEO provides screen instructions to proceed with the test.*

1. Put the cap on LEO gas outlet port (1 in Figure 26)

Note ► Use additional caps supplied in LEO's Hard case.

2. Put the cap on LEO gas inlet port with icon **IN** (2 in Figure 26).



Figure 26. Put caps on LEO ports.

3. Press on *Continue* (see Figure 27 a). The pump starts operating and aspirating air. The pressure inside LEO begins to rise. Wait until *aspiration* step is completed (see Figure 27 b). Figure 27 c appears.



Figure 27. How to set Pump Leak Test – aspiration step (a - b - c).

4. Put the cap on LEO gas inlet port with icon (see Figure 28) and press on *Continue* (see Figure 29 a). Wait *stabilization* time (see Figure 29 b), during this time the pump is not running. Wait until the *Leakage evaluation* step is completed (see Figure 29 c).



Figure 28. Put the cap on LEO gas inlet port.

📰 🚥 56% <mark>%</mark> 13:16	📲 56% 🚱 13:17	a= 56% 🖌 13:17
Pump Leak Test	Pump Leak Test	Pump Leak Test
To proceed, please put the provided cap on the "IN"	10% Wait 18 seconds for stabilization.	5% Wait 33 seconds for evaluation.
aspiration gas port. Then press on "Continue".	Pump is off.	Leakage is being checked.
Pressure 1241 mbar	Pressure 1261 mbar	Pressure 1182 mbar Leakage 0.02200 mbar+L/s
Abort Continue	Abort	Abort
а	b	С

Figure 29. How to set Pump Leak Test – stabilization and Leakage evaluation steps (a - b - c).

If the Leak test is passed, the pump works properly. The green label showing *OK* appears as in Figure 30.

📰 56% <mark>+</mark> 13:23				
Pump Leak Test				
ок				
Procedure performed successfully. No leakage found.				
Pressure 1132 mbar				
Leakage 0.00366 mbar × L / s				
Back				

Figure 30. Leak test successfully completed.

If the Leak test failed, a leakage in the pump is present and a red label showing *FAILED* appears. Make sure that caps are well inserted into LEO's ports and repeat the Leak test. If the Leak test failed again please contact Okolab support at support@oko-lab.com.

Tip \triangleright Okolab suggests to perform Leak test every time you replace a component inside LEO. You may be asked to run this test during remote assistance session.

9.3.4 Alarm

Press the *Alarm* icon (see Figure 31 a) to enter the *Alarm* subpage. You can set alarm parameters for each sensor by following the instructions below:

- 1. Select the sensor you want to set alarm parameters, for example CO2 (see Figure 31 b).
 - 2. Insert the Minimum (*Min*) and Maximum (*Max*) value within which the measurements can be considered valid (not in alarm) (see Figure 31 c). An alarm occurs when the reading value is out of the inserted values range.
 - 3. Press "Set" to save the alarm parameters.
 - 4. If you select the "Buzzer" checkbox, LEO triggers when it is in alarm condition (see Figure 31 b).



Figure 31. Alarm page (a – b - c).

5. Repeat the steps 1-4 to insert the alarm parameters for the variables on which you want to active an alarm.

Note ► *External sensor modules' icons are automatically disabled if sensor modules are not connected to LEO.*

9.3.5 System

Figure 32 b shows the System subpage. Press the *System* icon \times (see Figure 32 a) to see or modify system options:

- Date & Time. See paragraph 9.3.5.1.
- Power Saving. See paragraph 9.3.5.2.
- Memory. See paragraph 9.3.5.3.
- Info. See paragraph 9.3.5.4.



Figure 32. System page (a – b).

9.3.5.1 Date & Time

To set *Date & Time*, follow the instructions below:

1. Press the *Date & Time* icon (see Figure 33 a) to access the *Date & Time* page.

94% 🗐 13:06	49% 🖌 12:37
System	Date & Time
Date & Time Power saving Memory	Date: 19/07/2018 Charge
Info	Time: 12:37 Change
Home Settings View Device	Home Settings View Device
а	b

Figure 33. Date & Time configuration (a – b).

2. Press *Change* (see Figure 33 b) to enter the *Set Date* page.

Tip \blacktriangleright Okolab recommends to check the Date & Time Settings the first time you turn LEO on because the current Date & Time values are stored with every reading.

3. Change the date using + *and* – (1, 2, 3 in Figure 34 b).



Figure 34. Set Date (a – b).

4. Press Save (4 in Figure 34 b) to confirm.

5. Press *Change* (see Figure 35 a) to enter the *Set Time* page.

Note ► *The default time format is a 24-hours style clock.*

- 6. Change the time using + *and* (1 and 2 in Figure 35 b).
- 7. Select/deselect the 24 hours checkbox to have a 24 or 12 hours style clock. (3 in Figure 35 b).
- 8. Press *Save* to confirm (4 in Figure 35 b).



Figure 35. Set Time (a – b).

9.3.5.2 Power Saving

Press the *Power saving* icon \checkmark (see Figure 36 a) to set *Low brightness* feature and *Stand By* feature. *Low brightness* feature is enabled by default. Set the time period after which LEO will reduce the display's brightness by 15%.

Note ► *Display's brightness is automatically reduced when the remaining battery power is less than 5%.*

The *Stand By* feature is disabled by default. Enable the *Stand By* feature by flag the checkbox. Set the timeout period after which LEO's display switches off.

Press *Save* to confirm settings, see Figure 36.

Note ► The Stand By time range is 1-30 minutes.

Note ► *Press the power button to turn the display on.*



Figure 36. Power saving settings (a - b).

9.3.5.3 Memory

LEO stores up to 300 records. The memory shows how many records can be saved. Press the *Memory* icon (see Figure 37 a) to check the remaining space available in memory (see Figure 37 b).

Note ► *Before starting to measure, make sure that the space available on LEO memory is large enough to store new records.*



Figure 37. Memory check (a – b).

9.3.5.4 Info

Press the icon it access the System Information, as shown in Figure 38 a. This page contains the information related to LEO serial number and version (see Figure 38 b). Click on the right arrow icon to move to sensor module information panel.

Tip ► *Please have this information handy when contacting Okolab for support.*



Figure 38. System information (a – b).

9.3.6 Display

Press the *Display* icon to modify the display options (see Figure 39 a). As shown in Figure 39 b, in the *Display* page it is possible to set:

- *Brightness*: to modify the display brightness (see paragraph 9.3.6.1).
- *Theme*: to modify the theme (see paragraph 9.3.6.2).
- *Touchscreen*: to modify the touch options (see paragraph 9.3.6.3).



Figure 39. Display settings (a – b).

9.3.6.1 Brightness

To modify the display *Brightness*, press the icon \forall (see Figure 40 a) and use the slide bar displayed or simply press + *and* -, as shown in Figure 40 b. Press *Save* to confirm or *Cancel* to undo.



Figure 40. Display settings: Brightness (a – b).

Note ► *Display's brightness is automatically reduced when the remaining battery power is less than 5%.*

9.3.6.2 Theme

To modify the display *Theme*, press the icon *(see Figure 41 a)* and select the theme you prefer in the page that opens (see Figure 41 b).



Figure 41. Display settings: Theme (a – b).

9.3.6.3 Touchscreen

To change the touch sound frequency, press the *Touchscreen* icon (see Figure 42 a) and slide your finger along the *Buzzer frequency* bar (1 in Figure 42 b) or simply press + and -, then press *Save* to confirm or *Cancel* to undo.



Figure 42. Display settings: Touchscreen options (a – b).



9.3.7 Units

Press the *Units* icon (see Figure 43 a) to change the measurement units of temperature and pressure variables. There are three available options for both: temperature and pressure as shown in Figure 43 b. Select the measurement unit you prefer to use and press *Save* to confirm (3 in Figure 43 b).

Tip ► *The default measurement units are C° for temperature and mbar for pressure.*


Figure 43. Units (a – b)

9.3.8 Factory Reset

Press on *Factory Reset* icon **?** (see Figure 44 a) to restore the default factory settings, a pop-up dialog box appears (see Figure 44 b). Press *Ok* to start Factory Reset or *Cancel* to undo (see Figure 44 b).



Figure 44. Factory reset (a – b)

Note ► *Factory Reset will erase user calibration of all sensor modules connected.*

10 CO2-O2 Measurement process

LEO allows measurement of the CO2 and O2 concentration in two different modes:

- *Diffusion Mode,* without pump running. Use Diffusion Mode when the sampling gas has an overpressure higher than 10 mbarg (0.145 psig).
- *Note* ► *Do not exceed 300 mbar (4 psig) upstream of LEO*

Note ► LEO can operate at minimum flow rate of 30 ml/min and at maximum flow rate of 400 ml/min.

Tip ► *The optimal flow rate of the sampling gas is 150 ml/min.*

• *Aspiration Mode,* with pump running. Use Aspiration Mode when the sampling gas is at atmospheric pressure.

Tip \blacktriangleright The default value of the gas flow rate sucked by LEO is 150 ml/min. To change this value, see paragraph 9.3.3.

To start a gas measurement process, press the *ON/OFF* button and wait until the *Warming up time* ends. It will take 15 minutes for CO2-O2-MODULE to achieve stability. During this time, the message *"LEO is warming up"* appears (1 in Figure 45).



Figure 45 . LEO is warming up.



CO2 and O2 gas concentration measurement in Diffusion Mode cannot occur simultaneously with a CO2 – O2 gas concentration measurement in Aspiration Mode.

10.1 CO2-O2 Measurement process: Preliminary Operations

Before starting the CO2-O2 Measurement Process, you have to follow some preliminary operations. See paragraph 10.1.1 for the CO2-O2 measurement in Diffusion Mode or paragraph 10.1.2 for the CO2-O2 measurement in Aspiration Mode.

10.1.1 Diffusion Mode – Preliminary Operations

- 1. Connect the **clear connector** of the Tube-A to LEO gas input port labeled **IN** (1 in Figure 46, Configuration a and in Figure 46, configuration b).
- Put the cap on LEO gas input port labeled with icon (2 in Figure 46, Configuration a and in Figure 46, configuration b)
 - 3. Connect the **red connector** of the Tube-A to the **green connector** of Tube-B (3 in Figure 46, Configuration a).

If the sampling gas is humid, connect the **red connector** of Tube-A (3 in Figure 46, Configuration b) and the **green connector** of Tube-B (4 in Figure 46, Configuration b) to the Moisture Trap. In this case, operate in *Wet Mode* (see paragraph 10.1.5)

- 4. Connect the end of Tube-B to the incubator or to the point where measurement is required (4 in Figure 46, Configuration a and 5 in Figure 46, Configuration b)
- 5. Okolab recommends the use of a purge tube attached to LEO gas outlet port (5 in Figure 46, Configuration a and 6 in Figure 46, Configuration b). You can use the supplied additional tube as tube for purging. Use the supplied female luer connector to connect the additional tube to LEO gas outlet port. The purge tube has to be placed in a well ventilated environment.

Tip \blacktriangleright Use the fitting tubes and additional tubes in order to allow the compatibility of the supplied tubing with the sampling gas port of your incubators.



Figure 46. LEO Connections in Diffusion Mode (Configuration a - Configuration b).

6. Press the *Pump/Diffusion Mode* button (see Figure 47 a) to select the Diffusion Mode (see Figure 47 b).

Once you have followed the step mentioned above, LEO is ready to be used in Diffusion Mode. See paragraph 10.1.3 to start a Single Point Measurement or paragraph 10.1.4 to start a Gas Logging.



Figure 47. How to select the Diffusion Mode (a – b).

10.1.2 Aspiration Mode – Preliminary Operations

- 1. Connect the **clear connector** of the Tube-A to LEO gas input port labeled (1 in Figure 48, Configuration a and in Figure 48, Configuration b).
 - 2. Put the cap on LEO gas input port labeled with icon **IN** (2 in Figure 48, Configuration a in Figure 48, Configuration b).
 - 3. Connect the **red connector** of the Tube-A to the **green connector** of Tube-B (3 in Figure 48, Configuration a).

If the sampling gas is humid, connect the **red connector** of Tube-A (3 in Figure 48, Configuration b) and the **green connector** of Tube-B (4 in Figure 48, Configuration b) to the Moisture Trap. In this case, operate in *Wet Mode* (see paragraph 10.1.5)

- 4. Connect the end of Tube-B to the incubator or to the point where measurement is required (4 in Figure 48, Configuration a and 5 in Figure 48, Configuration b)
- 5. Okolab recommends the use of a purge tube attached to LEO gas outlet port (5 in Figure 48, Configuration a and 6 in Figure 48, Configuration b). You can use the supplied additional tube as tube for purging. Use the supplied female luer connector to connect the additional tube to LEO gas outlet port. The purge tube has to be placed in a well ventilated environment.

Tip \blacktriangleright Use the fitting tubes and additional tubes in order to allow the compatibility of the supplied tubing with the sampling gas port of your incubators.



Figure 48. LEO Connections in Aspiration Mode (Configuration a - Configuration b).

6. Press the Pump/Diffusion Mode button (see Figure 49 a) to select the Aspiration Mode (see Figure 49 b).

Once you have followed the step above, LEO is ready to be used in Aspiration Mode. See paragraph 10.1.3 to start a Single Point Measurement or 10.1.4 to start a gas measurements logging.



Figure 49. How to select the Aspiration Mode (a – b).

10.1.3 CO2-O2 Single Point Measurement

To start a single point measurement, follow the instructions below:

1. Select the Device for which the data will be stored (see paragraph 9.3.1).

Note \blacktriangleright If no device is selected, the data will be stored on the current device shown on the Homepage (1 in Figure 50 a).

- 2. Press the CO2-O2 Single Point Measurement icon **(see Figure 50 a)**. A pop-up dialog box appears (see Figure 50 b). Select *Auto* for an automatic or *Manual* for a manual measurement.
 - Auto: the CO2-O2 Single Point measurement terminates automatically when the gas reading value becomes stable.
 - Manual: you have to click again on the CO2-O2 Single Point Measurement Icon to stop the measurement.



Okolab recommends using the Auto measurement only when the gas flowrate sent to LEO is higher than 130 ml/min.

Tip ► Okolab suggests to use the Manual measurement to meet fully accuracy requirements.



Figure 50. CO2-O2 single point measurement (a – b - c).

During the CO2-O2 Single Point Measurement, CO2 and O2 gas concentration values appear on the Homepage. The *CO2-O2 Single Point Measurement* icon becomes **(see Figure 50 c)**.

3. Press the *View* icon to view data stored (see Figure 51 a). More details are reported in chapter 12.



Figure 51. View data stored (a – b - c).

10.1.4 CO2-O2 Logging

To start a CO2-O2 Logging follow the instructions below.

1. Select the Device for which the data have to be stored (see paragraph 9.3.1).

Note \blacktriangleright If no device is selected, the data are stored on the current device shown on the Homepage (1 in Figure 52 a).

- 2. Press the CO2-O2 Logging icon 📫 (see Figure 52 a). The CO2-O2 Logging Setting is shown.
- 3. Insert the measurement times using + and (1, 2 and 3 in Figure 52 b):
 - **Each:** the time between two measurement points. It is the acquisition frequency. The time can be inserted in minutes and seconds.
 - For: the time interval of the CO2-O2 Logging. The time can be inserted in hours and minutes.
 - **Pump Time:** the time, in minutes, the pump is active and aspirates gas, prior to saving a reading. This parameter is available only if the Aspiration mode is selected.
- 4. Enable/Disable the alarm on the CO2 and O2 concentration (4 in Figure 52 b). Then press *Start* to start the measurement (5 in Figure 52 b).

Note \blacktriangleright If CO2 and/or O2 check boxes are enabled (see Figure 52 b), the alarm triggers when the reading concentrations values are out of the set range. See paragraph 9.3.4 to set the gas alarm range.

Note ► *The Pump Run Time CANNOT be less than 1 min.*

Note \blacktriangleright If the time inserted in Each is less than the Pump Time, the Gas Logging occurs with the pump continually ON. Each time can be set at a minimum of 5 seconds.

Tip ► *The recommended Pump Time is 3 min.*



Figure 52. Gas logging (a - b - c).

When CO2-O2 Logging is in progress, CO2 and O2 gas concentration measurement values appear on the Homepage. The CO2-O2 Logging icon becomes

Note ► During a CO2-O2 Logging, the timer (2 in Figure 52 c) indicates the remaining time for completing the time interval, set as "For" value (2 in Figure 52 b).

When CO2-O2 Logging is completed or if you stop it by clicking on the icon (see Figure 52 c), the last values remain on the Homepage for 10 seconds, or until a new measurement occurs.

5. Press the *View* icon (see Figure 51 a) to view data stored. More details are reported in chapter 12.

10.1.5 Wet Mode

LEO measures the sampled gas composition at the environmental pressure, temperature and humidity conditions. If the gas sample is very humid, you must use the moisture trap between the incubator and LEO (see paragraph 5.2 and paragraph 10.1), the loss of water molecules changes the composition of the gas and has an effect on the CO2 – O2 measurement. To address this issue and correct the CO2 and O2 measurement to account for the incubator real condition, you can use the *Wet Mode*.

The *Wet Mode* is a LEO feature allowing you to set the incubator temperature and relative humidity values. LEO will correct the CO2 and O2 values by accounting for the real water content of the gas drawn from the incubator.

In order to set *Wet Mode*, press the *Wet/Dry Mode* button (see Figure 53 a). A pop-up dialog box appears. Insert the temperature and relative humidity values of the Wet Incubator and then press *Save*, as shown in Figure 53 b.



Figure 53. How to select Wet Mode (a – b).

11 Temperature Measurement Process

The temperature measurement process requires a T1 or T2-modules connected to LEO.

The T1-MODULE is generally used if the diameter of the temperature monitoring port is smaller than 2mm. The T2-MODULE is generally used for diameters larger than 2mm.

11.1 T Modules - Preliminary Operations

Follow the instructions below to connect T1 and T2-modules to LEO.

- 1. Connect T1-MODULE (or T2-MODULE) to one of the probe connectors on the top of LEO, see Figure 54 a. The reading temperature value is immediately shown.
 - 2. Connect T2-MODULE (or T1-MODULE) to the other probe connector (for a simultaneous T1 and T2 temperature measurement), see Figure 54 b. The reading temperature value is immediately shown.
 - 3. Introduce the T1 probe (or T2 probe) into the temperature monitoring port of the incubator.
 - 4. Wait for LEO starts reading a temperature value. The reading is shown on the homepage (to the right of T1 or T2).

Note ► *The two temperature connection ports can be used both for T1 and T2 modules (see Figure 54 a and b).*

Note \triangleright T1-MODULE and T2-MODULE can be used simultaneously. They can also be used during a gas measurement in Diffusion Mode or in Aspiration Mode.



Figure 54. T1 and T2 connections (a – b).

11.2 Temperature Single Point Measurement

After connecting the temperature module (T1 or T2-MODULE), follow the instructions below to start a temperature Single Point Measurement.

1. Select the Device for which data has to be stored (see paragraph 9.3.1).

Note \blacktriangleright If no device is selected, the data is stored on the current device shown on the home page (1 in Figure 55 a).

Tip ► During a gas measurement, you can start a temperature measurement and select a different Device for which temperature data shall be stored.

- 2. Wait until the temperature reading becomes stable.
- 3. Press the Temperature Single Point Measurement icon **to** (see Figure 55 a).
- 4. Press the View icon to view stored data (see Figure 55 b). More details are reported in chapter 12.



Figure 55. Homepage during a temperature single point measurement.



Before clicking on Single Point Temperature Measurement, wait for reading temperature is stable.

11.3 Temperature Logging

To start a temperature logging follow the instructions below:

1. Select the Device for which the data have to be stored (see paragraph 9.3.1).

Note \blacktriangleright If no device is selected, the data are stored on the current device shown on the homepage (1 in Figure 56 a).

Tip ► During a gas measurement, you can start a temperature measurement and select a different Device for which temperature data shall be stored.

- 2. Wait for the temperature reading is stable.
- 3. Press the *Temperature Logging* icon (see Figure 56 a). The *Temperature Logging* settings page is shown.
- 4. Insert the measurement time using + *and* (see Figure 56 b):
 - **Each:** the time between two measurement points. It is the acquisition frequency. The time can be inserted in minutes and seconds. Each time can be set at minimum of 5 seconds.
 - For: the time interval of the Temperature Logging. The time can be inserted in hours and minutes.
- 5. Enable/Disable the alarm on T1 and/or T2 (1 in Figure 56 c) and then press *Start* to start the measurement (2 in in Figure 56 c).

Note \blacktriangleright If T1 and/or T2 check boxes are enabled (see Figure 56 c), the alarm triggers when the reading temperature values are out of the set range. See paragraph 9.3.4 to set the temperature alarm range.



Figure 56. Temperature logging (a - b - c)

When the Temperature Logging is in progress, the *Temperature Logging* icon becomes 📫 (see Figure 57

b).

Note \triangleright During a Temperature Logging, the timer (1 in Figure 57 a) indicates the remaining time for completing the time interval, set as "For" value (see Figure 56 b).

Note \blacktriangleright The Temperature Logging can be interrupted before the time set by clicking on the icon \bowtie (see Figure 57 a).



Figure 57. Temperature logging: view data

Press the View icon to view the data stored (see Figure 57 b). More details are reported in chapter 12.

12 Reading Data

LEO allows storing sensors' measurements and then to view (see paragraph 12.1), download (see paragraph 12.2) or delete (see paragraph 12.3) the stored data.

12.1 Viewing Data

To view the collected Data, please follow the instructions below.

- 1. Click the *View* icon (see Figure 58 a) to see the *View Data* (see Figure 58 b). The values in this page are the last measurement of each sensor module.
 - 2. Use the UP and DOWN arrows to scroll the device list (1 in Figure 58 b).
 - 3. Use the LEFT and RIGHT arrows to view the measurements by date (2 in Figure 58 b).
 - 4. Press the *icon* (3 in Figure 58 b) to select the variables to see, download or delete. Flag the variables and then press *OK* button to save, see Figure 58 c.

Note ► You can display up to 4 variables at any one time.



Figure 58. How to access to view data (a - b - c).

5. Press the device name (for example, the *incub1*) (see Figure 59 a). The page with all values stored in that date for the selected device opens (see Figure 59 b).



Figure 59. View Data (a - b - c).

Each line corresponds to a reading and the reading type is identified by the prefix (1 in Figure 60):

- *GAS*, if the reading refers to a CO2-O2 gas concentration measurement.
- *T1/T2*, if the reading refers to a temperature measurement with T1 and/or T2-MODULE (only if you used a T1 and or T2-MODULE purchased separately).

In addition to the measurement type, LEO also shows the number of the measurements (3 in Figure 60) for each measurement.

The number of the measurements depends on whether you started a single point measurement or a gas/temperature logging:

- For single point measurement, LEO shows [1] (3 in Figure 60) and the time when the measurement is over (2 in Figure 60)
- For a gas/temperature logging, LEO shows a number that depends on the values inserted for *Each* and *For* parameters before starting the measurement (see paragraph 10.1.4 and 11.3) and measurement start time.



Figure 60. Data stored format.

6. To display measurement details, press on a line (see Figure 59 b). The page shown in Figure 59 c opens. If data refer to a gas or temperature logging, they can be seen both in table format and in graph format.

- 7. Use the DOBLE UP and DOUBLE DOWN arrows (1 in Figure 59 c) to scroll more quickly the measurement list.
- 8. Press the *Info* icon (2 in Figure 59 c) to display the measure's details (see Figure 61)



Figure 61. Measure details

Tip \blacktriangleright To view the data in graph format, press on icon \checkmark (3 in Figure 59 c).

If you have inserted the alarm values in the relative page before starting the measurement (see paragraph 9.3.4), they are shown in the top right of the page (4 in Figure 59 c). In the example reported in Figure 59 c, the CO2 alarm parameters, set before starting the measurement, are 1.0 as CO2 minimum (*Min*) and 7.2 as CO2 maximum (*Max*) values.

12.2 Downloading Data

LEO is equipped with local memory to store sensor modules' readings. These data can be downloaded on a USB flash drive on user request. To download the stored data of all devices, stored in a precise time interval, follow the instructions below:

1. Connect the USB key to the MINI-USB port as shown in Figure 62. The *Usb* Icon appears on the Homepage (1 in Figure 63).



Figure 62. Download Data.



Figure 63. USB Icon displayed when a USB key is connected to LEO.

- 2. Press the View icon to open the *View Data* page (see Figure 64 a).
- 3. Press the icon 🔽 to access to the *Download/Delete Data* page (see Figure 64 b).
- 4. Insert the start-time and the end-time corresponding to the data interval to download (1 and 2 in Figure 64 c).
- 5. Enable the *Download* check box to download the selected data (3 in Figure 64 c). Then press *OK* to download data to the USB key (4 in Figure 64 c).



Figure 64. How to Download Data (a - b - c).

Tip \blacktriangleright To download all the data stored on LEO internal memory, flag All checkbox (1 in Figure 65) and Download checkbox (2 in Figure 65). Then press OK to confirm (3 in Figure 65).

📰 83% <mark>%</mark> 12:41	l
Download/Delete Data	
1	
- 10	
_2 _	
Download	
Cancel OK 3)
Home Settings View Device	

Figure 65. How to download all data stored on LEO.

To download all the readings of a single device, connect the USB key to the MINI-USB port (see Figure 62) and follow the instructions below:

- 1. Press the *View* icon to open the View Data page (see Figure 64 a).
 - 2. Press the device name (see Figure 66 a).
 - 3. Press the icon 🕤 (see Figure 66 b) in the page that opens. A pop-up dialog box appears.

4. Enable the Download check box (1 in Figure 66 c) and then press *OK* to download the selected data to the USB Key (2 in Figure 66 c).



Figure 66. How to Download Data for a single device (a - b - c).

To download a single reading of a single device, connect the USB key to the MINI-USB port (see Figure 62) and follow the instructions below:

- 1. Press the device name (see Figure 59 a).
- 2. Press a single reading (see Figure 59 b) and then press the icon 🐨 (see Figure 67 a) in the page that opens. A pop-up dialog box appears.
- 3. Enable the *Download* check box (1 in Figure 67 a) and then press OK to download the selected data to the USB Key (2 in Figure 67 b).

54% 🚱 06:03 P	M 📲 81% 🚱 11:32
incub1 - 6 Feb 2018	Default - 23 Jul 2018 📑
GAS	GAS dry 02 alarms: 2.6 - 9.8
CO2 O2 CO2 p O2 p	CO2 O2 pCO2 pO2
% % mbar mbar	% % mbar mbar
18:01:01 6.03 5.02 61 50	11.04.02 010 10.05 1 100
18:01:03 5.95 5.00 60 50	Download/Delete
18:01:05 5.97 5.02 60 50	11:06:23 0.18 19.96 1 199
18:01:07 6.03 5.00 61 50	11:07:24
18:01:09 6.02 4.98 60 50	Download 🗸 📕 elete
18:01:11 6.04 5.04 61 51	
18:01:13 6.03 4.95 61 50	
18:01:15 6.02 4.95 60 50	Cancel
18:01:17 5.99 5.02 60 50	
18:01:19 6.02 4.97 60 50	
	2
Home Settings View Device	Home Settings View Device
а	b

Figure 67. How to Download Data for a single reading (a - b).

 $Tip \triangleright$ The data will be downloaded in the USB drive as shown in Figure 68.

iie Home	Share Vi	Drive Tools ew Manage	OKOLAB (K:)				
Pin to Quick Copy access	Paste Cop Poste	y path te shortcut	Copy to · Organize	New item •	Properties Open Open	Select all Select none Invert selection Select	
← → * ↑ =	> This PC	OKOLAB (K:)					
	^	Name	^	Date modified	Type Siz	e :	
A Quick access		Default_2018	-09-19_10-26-11_T.txt	19/09/2018 10:49	Text Document	1 KB	
Desktop	*	Default 2018	-09-19 10-27-15 T.txt	19/09/2018 10:49	Text Document	1 KB	
👆 Downloads	1	Default 2018	-09-19 10-27-50 T.txt	19/09/2018 10:49	Text Document	1 KB	
Documents	*	Default_2018	-09-19_10-35-35_GAS.txt	19/09/2018 10:49	Text Document	1 KB	

Figure 68. Downloaded files on the pen drive

Tip ► *Please, use Microsoft Excel to open the downloaded .txt files.*

You can download LEO DATA IMPORT Excel Macro from Okolab website <u>www.oko-lab.com</u>. LEO DATA IMPORT Excel Macro helps you to organize your stored data.



Figure 69. Leo Data Import Excel Data

Press *Select folder* and choose the folder that contains LEO data. LEO Data Import Excel Macro automatically creates a new Excel file in the same folder with LEO data. The new excel file contains all LEO data automatically organized.

12.3 Deleting Data

To delete the stored data, follow the instructions below.

- 1. Press the *View* icon (see Figure 70 a), the *View Data* page opens (see Figure 70 b).
 - 2. Press the icon (see Figure 70 b) to access the *Download/Delete* page.
 - 3. Insert the start-time and the end-time corresponding to the data interval to delete (1 and 2 in Figure 70 c).
 - 4. Enable the *Delete* check box to delete the selected data (3 in Figure 70 c). Then press *OK* button to delete data (4 in Figure 70 c).



Figure 70. How to Delete Data (a - b - c).

Tip \blacktriangleright To delete all the data stored on LEO internal memory, flag All checkbox (1 in Figure 71) and Delete checkbox (2 in Figure 71). Then press OK to confirm (3 in Figure 71).



Figure 71. How to Delete all data stored on LEO.

To delete all the readings of a single device follow the instructions below:

- 1. Press the *View* icon to open the View Data page (see Figure 70 a).
- 2. Press the device name (see Figure 72 a).
- 3. Press the icon 🐨 (see Figure 72 b) in the page that opens. A pop-up dialog box appears.
- 4. Enable the *Delete* check box (1 in Figure 72 c) and then press *OK* to download the selected data to the USB Key. (2 in Figure 72 c).



Figure 72. How to Delete Data for a single device (a - b - c).

To delete a single reading of a single device follow the instructions below:

- 1. Press the device name (see Figure 72 a).
- 2. Press a single reading (see Figure 73 a) and then press the icon 🕤 in the page that opens (see Figure 73 b). A pop-up dialog box appears.
- 3. Enable the *Delete* check box (1 in Figure 73 c) and then press *OK* to delete the selected data (2 in Figure 73 b).



Figure 73. How to Download Data for a single reading (a - b).

Tip \blacktriangleright You can download and delete simultaneously collected data by enabling both Download and Delete checkboxes, as shown in Figure 74.



Figure 74. How to Delete and Download Data simultaneously.

13 CO2-O2-MODULE Calibration

Press on *Settings* icon ^(C) to access to the *Settings* subpage (see Figure 75 a). Press on *Calibration* icon

(see Figure 75 b) and select CO2/O2 icon (see Figure 75 c). The CO2/O2 Calibration page appears as in Figure 76).



Figure 75. How to set CO2/O2 Calibration (a - b - c)



Figure 76. CO2/O2 Calibration page.

CO2/O2 Calibration page contains five subpages:

- *View Current*: to view factory calibration or CO2/O2 User Calibration data. See paragraph 13.1.
- *Span Calibr.* : to run CO2/O2 User Calibration using certified span gas. See paragraph 13.2.

Note ► *Span gas is a gas mixture of precisely defined composition used as reference gas in the calibration of gas meters. Calibration is an important issue for all analytical instruments and is done by exposing the sensor*

to a gas of known concentration used as a comparative standard. The calibration procedure detects and deletes the reading error.

- *Factory*: to cancel the last CO2/O2 User Calibration and restore the factory values. See paragraph 13.3.
- *CO2 Zero Reset:* to correct the zero of the CO2 sensor. See paragraph 13.4.
- *Offset: to* set CO2 and/or O2 offset using an external gas meter. See paragraph 13.5.

13.1 View Current

LEO is equipped with a factory calibrated CO2-O2-MODULE using certified gas cylinders. The factory calibration certificate is included in electronic format and can be seen in the *CO2/O2 Calibration* page. Press the

View Current icon (see Figure 77 a) to view the factory calibration or CO2/O2 User Calibration data. If no CO2/O2 User Calibration has been made, there are only the *CO2* and/or O2 *Factory Calibration* subpages, as in the example shown in Figure 77 b and c.

📰 56% 🔒 13:2	26	82% 🖌 10:25	and the second s	82%	6 😽 🛛 10:24
CO2/O2 Calibration	View Calibr	ation	Vi Vi	ew Calibrati	on
	CO2 Factory Calibra	ation 🗪	O2 Factory	Calibration	
View current Span Calibr. Factory	Operator Calibration Date	okolab 17/05/2018	Operator Calibration Date		okolab 17/05/2018
,	Certified % Reading %	6 Deviation %	Certified %	Reading %	Deviation
CO	0.00 0.00	0.00	0.00	0.00	0.00
	6.02 6.02	0.00	4.98	4.99	0.01
Zero Beset Offset	9.95 10.02	0.07	10.05	10.07	0.02
	19.10 19.27	0.17	20.99	21.05	0.06
		1		☆ ■	
Home Settings View Device	Home Settings	/iew Device	Home Se	ttings View	Device
а	b			C	

Figure 77. CO2/O2 View Calibration (a – b – c).

The recommended CO2-O2-MODULE calibration interval is six months. However, Okolab suggests to verify LEO's reading each month or before each important measurement by employing a certified span gas. To achieve the best accuracy, perform the gas measurement immediately after the CO2/O2 User Calibration.

13.2 Span Calibration

To improve LEO reading accuracy, it is possible to perform CO2/O2 User Calibration using span gases.

If you want to achieve the best accuracy in the sensors' measurement range 0 – 10%, Okolab suggests to use a certified span gas cylinder with composition similar to the gas used during your working standard conditions. In this case, after the CO2/O2 User Calibration you can achieve an accuracy of $\pm 0.1\%$.

If you want to achieve the best accuracy in the sensors' full measurement range, Okolab suggests to use up to two span gases:

• Certified span gas cylinder with composition similar to the gas used during the standard working conditions

• Certified span gas cylinder with higher composition (usually 19% CO2 and 21% O2) to perform two-point calibration together with previous tank.

The certified gas should have a relative uncertainty lower than 1.0%. In general, the span gas cylinders should present a CO2 concentration higher than 1% and lower than 19%, an O2 concentration higher that 1% and lower than 24%.

Before starting CO2/O2 User Calibration, it is necessary to execute the Preliminary Operations, as described in the subsequent paragraphs.

Note ► Okolab recommends to perform the CO2 Zero Reset procedure before each CO2/O2 User Calibration (see paragraph 13.4).

13.2.1 CO2/O2 User Calibration using Span Gas Cylinder: Preliminary Operations

13.2.1.1 Span Gas Cylinder with Pressure Regulator

Calibration Kit and Tube-A are required to assure the correct gas flow rate. Follow the instructions below to connect LEO to the span gas cylinder equipped whit a pressure regulator:

- 1. Connect the **clear connector** of Tube-A to LEO gas inlet port with icon **IN** (1 in Figure 79)
- 2. Put the cap on LEO gas inlet port with icon = (2 in Figure 79).
- 3. Connect the **clear connector** of the Calibration Kit to **red connector** of Tube-A (3 in Figure 79)



Figure 78. Calibration Kit connected to Tube-A

- 4. Connect end of the Calibration Kit to the gas cylinder (4 in Figure 79).
- 5. Okolab recommends the use of a purge tube attached to LEO gas outlet port (5 in Figure 58). The purge tube has to be placed in a well ventilated environment. You can use the supplied additional tube as tube for purging.
- 6. Set 1.0 barg (14.5 psig) on the pressure regulator of the gas cylinder (6 in Figure 58).

Note ► Do not exceed 1.5 barg (22 psig).

Tip \blacktriangleright Use the fitting tubes and additional tubes in order to allow the compatibility of the supplied tubing with the gas port of your gas cylinders.

Note ► The Moisture trap must not be used during the CO2/O2 User Calibration.



Figure 79. How to connect LEO to the gas cylinder with pressure regulator.

13.2.1.2 Span Gas Cylinder with Flow Regulator

Tube-A and Tube-B are required to connect LEO to a span gas cylinder equipped with a flow regulator. Follow the instructions below to connect LEO to the gas cylinder with flow regulator:

- 1. Connect the **clear connector** of Tube-A to LEO gas inlet port labeled with icon **IN** (1 in Figure 59).
 - 2. Put the cap on LEO gas inlet port labeled with icon (2 in Figure 59).
 - 3. Connect the red connector of the Tube-A to the green connector of Tube-B (3 in Figure 59)
- 4. Connect the end of Tube-B to the span gas cylinder regulator (4 in Figure 59).

Tip \blacktriangleright Use the fitting tubes or additional tubes in order to allow the compatibility of the supplied tubing with the gas port of your gas cylinders.

- 5. Okolab recommends the use of a purge tube attached to LEO gas outlet port (5 in Figure 59). The purge tube has to be placed in a well ventilated environment. You can use the supplied additional tube as tube for purging.
- Open the span gas cylinder regulator by using the valve (6 in Figure 80). Set the flow rate in the range 0.1 -0.3 l/min. Do not exceed the maximum flow rate of 0.4 l/min.



Figure 80. How to connect LEO to the gas cylinder with flow regulator.

13.2.2 CO2/O2 User Calibration with Certified Gas at atmospheric pressure: Preliminary Operations

If you use a gas at atmospheric pressure for CO2/O2 User Calibration, you can calibrate LEO in Aspiration Mode by using the internal Pump.

Figure 79 shows how to connect LEO during the CO2/O2 User calibration with a gas at atmospheric pressure by using the Tube-A. Follow the instructions below to connect LEO to Tube-A:

- Connect the clear connector (a) of the Tube-A to LEO gas inlet port labeled with icon (1 in Figure 81).
 - 2. Put the cap on LEO gas inlet port labeled with icon **IN** (2 in Figure 81).
 - 3. Connect the **red connector** (b) of Tube-A to the gas reference at atmospheric pressure (1 in Figure 81).
 - 4. Okolab recommends the use of a purge tube attached to LEO gas outlet port (5 in Figure 79). The purge tube has to be placed in a well ventilated environment. You can use the supplied additional tube as tube for purging.



Figure 81. How to connect LEO during a calibration in Aspiration Mode.

13.2.3 Span Setting

Press *Span Calibration* icon ut to access to the *Span Calibration* submenu (see Figure 82 a).



Figure 82. Span gas setting page (a – b).

To set all parameters of the span gas used to calibrate the CO2-O2-MODULE follow the instructions below:

1. Press *Span Setting* icon *in* the page that opens (see Figure 82 b) to access to the *Span Settings* page.

Make sure to set the correct parameters of the reference span gas used to calibrate the CO2-O2-MODULE in the *Span Settings* page (see Figure 83 a).

Tip ► *LEO* can be calibrated for CO2 sensor, O2 sensor or both.

gases used for the calibration procedure.

Tip For example, the Figure 83 a shows the settings for a span calibration by two span gases (Span Gas 1 and Span Gas 3); during the procedure the Span Gas 1 will be used for calibrating both sensors (CO2 at 6.00 % and O2 at 5.00 %), the Span Gas 3 only for calibrating the CO2 sensor (at 19 %).



Figure 83. Span Settings page (a – b).

- 3. If you use a gas with an overpressure for the CO2/O2 User Calibration, you can calibrate LEO in *Diffusion Mode*, as shown in Figure 83 b.
- 4. If you use a gas at atmospheric pressure for the CO2/O2 User Calibration, you can calibrate LEO in *Aspiration Mode* by pressing on icon as shown in Figure 84 a. The Pump indication appears (see Figure 84 b) and you can press on *Save* to confirm.

Note ► Okolab recommends to use 150 ml/min as pump flowrate value during a calibration in Aspiration Mode. See paragraph 9.3.3 to change the pump flowrate value.



Figure 84. Span Setting in Aspiration mode (a - b).

13.2.4 Start

After checking the reference gas concentrations in Span Settings page (see Paragraph 13.2.3), press the

Start icon (see Figure 85 a) to start a Span Calibration. The pop-up dialog shown in Figure 85 b appears. Select *Auto* for an automatic calibration or *Manual* for a manual calibration.



Figure 85. Span gas start (a – b)

13.2.4.1 Auto Gas calibration

By pressing on Auto in Figure 85 b, Figure 86 a appears.

Press *Continue* (see Figure 86 a) to start the Span Calibration after checked the proper span gas cylinder is connected to LEO.

During an Auto calibration, you can see CO2 and O2 readings on the display (see Figure 86 b).

Wait until CO2/O2 User Calibration is completed for the first span gas and the message shown in Figure 86 c appears. CO2/O2 User Calibration stops automatically, when the gas measurement becomes stable.

Then press *Continue* to start the next step of the calibration with Span Gas 2 and/or Span Gas 3, as shown in Figure 86 c (after connecting the span gas to use in the next step).



Figure 86. How to set Auto Gas Calibration (a – b - c).

At the end of the calibration procedure, CO2 and O2 offset values are shown as in Figure 87. Press *Save* to save the offsets or *Cancel* to undo.

72% 🚱 11:07
Gas Calibration
Calibration completed
SPAN 1
CO2 offset: 0.2 @ 6.00 O2 offset: -0.4 @ 5.00
SPAN 2
SPAN 3
CO2 offset: 0.44 @ 19.00 -
Save

Figure 87. Auto Gas Calibration completed.

13.2.4.2 Manual Gas Calibration

By pressing on *Manual* in Figure 85 b, Figure 88 a appears. Press *Continue* (see Figure 88 a) to start the Span Calibration after checking that the proper span gas cylinder is connected to LEO.

During a Manual calibration, you can see the CO2 and O2 readings on the display.

Wait until the CO2 and O2 readings became stable.

Tip ► Okolab suggests waiting at least 10 minutes before stopping Manual Calibration, if using Okolab Calibration Kit.

When the CO2 and O2 readings are stable press Set as shown in Figure 88 b.

When Span Gas 1 is completed the message shown in Figure 88 c appears. If you have Span Gas 2 and/or Span Gas 3, you have to connect Span Gas 2 / 3 bottle, then press on *Continue*.

When calibration is completed, CO2 and O2 offset values are shown as in Figure 87. Then press *Save* to save the offsets or *Cancel* to undo.



Figure 88. How to set Manual Gas Calibration (a - b - c).

View Current icon to view the CO2/O2 User Calibration data.

13.3 Factory

LEO allows to delete the last CO2/O2 User Calibration and to restore the factory values. To reset the current sensors calibration:

- 5. Press the Factory icon \bigcirc (see Figure 89 a).
- 6. Flag the checkbox of the variables for which you want to delete the CO2/O2 User Calibration (see Figure 89 b).

Tip \blacktriangleright *The reset can be done on CO2 and O2 sensors contemporary or only on CO2 or O2 sensor.*

7. Press *Reset* to confirm or *Cancel* to undo.



Figure 89. Factory Calibration (a – b)

13.4 CO2 Zero Reset

LEO allows adjusting the zero of the CO2 sensor using the CO2 Zero Reset procedure.

The first *CO2 Zero Reset* procedure is performed at our factory. To improve LEO reading accuracy the *CO2 Zero Reset* should be repeated every thirty days. LEO will remind you every thirty days to perform the *CO2 Zero Rest* before the CO2/O2 User Calibration and before a CO2 single or logging measurement (see Figure 90 b).



Figure 90. LEO warning messages suggesting the CO2 Zero Reset (a - b).

Note ► *After thirty days from the last CO2 Zero Reset, you must perform the CO2 Zero Rest before the CO2/O2 User Calibration.*

Note ► *After thirty days from the last CO2 Zero Reset, repeating the CO2 Zero Reset is highly recommended but not mandatory before a gas measurement.*

Press on *CO2 Zero Reset* icon (see Figure 91) to correct the zero of the CO2 sensor, the pop-up in Figure 91 b appears.

You can decide to use Air or N2 for the CO2 Zero Reset procedure.



Tip ► Okolab suggests to start the CO2 Zero Reset using N2, use Air only if N2 is not available.

Figure 91. CO2 Zero Reset (a – b).

13.4.1 CO2 Zero Reset with Air

Press on Air to start the CO2 Zero Reset if you are using air. See Figure 92 a.



Figure 92. CO2 Zero Reset with Air (a - b - c).

Pressing on *Air*, LEO reminds you to connect the Soda Lime kit, Figure 92 b. To connect the Soda Lime Kit to LEO follow the instructions below:

- 1. Connect the **clear connector** of Tube-A to LEO gas inlet port labeled with icon = (1 in Figure 94).
- 2. Put the cap on LEO gas inlet port labeled **IN** (2 in Figure 94).
3. Connect the clear connector of Soda Lime Kit to the red connector of Tube-A (3 in Figure 94)



Figure 93. Soda Lime Kit connected to Tube-A.

4. Press *Ok* (see Figure 92 b). A pop-up with the procedure progress appears, see Figure 92 c.

Tip ► *The Soda Lime kit removes acid gases, and CO2 from air.*

Note ► *The Soda Lime kit has to be replaced when the Soda Lime filter changes color turning purple. Contact Okolab to purchase a new Soda Lime kit.*



Figure 94. How to connect LEO during a CO2 Zero Reset procedure with Air.

The *CO2 Zero Reset* procedure takes 15 minutes, the remaining time is shown on the display during the procedure (see Figure 92 c).

At the end of the *CO2 Zero Reset* procedure with Air, a pop-up message is shown as in Figure 95. Press *Ok* to return to the *CO2/O2 Calibration* page.



Figure 95. CO2 Zero Reset procedure with Air completed.

If the *CO2 Zero Reset* procedure has failed the message shown in Figure 96 appears. In this case Okolab recommends to repeat the *CO2 Zero Reset* procedure and if it fails again please contact Okolab support at support@oko-lab.com.



Figure 96. CO2 Zero Reset procedure with Air has failed.

13.4.2 CO2 Zero Reset with N2

To start the *CO2 Zero Reset* procedure using N2, press on *N2* as shown in Figure 97 a. Pressing on *N2*, LEO reminds you to connect a Nitrogen bottle at 1.0 barg (14.5 psig) to LEO. See Figure 97 b.



Figure 97. CO2 Zero Reset on N2 (a - b - c).

Figure 99 shows how to connect LEO to N2 cylinder equipped with a pressure regulator. Note that the Calibration Kit and Tube-A are required to assure the correct gas flow rate. Follow the instructions below to connect LEO to the N2 cylinder with pressure regulator:

- 1. Connect the **clear connector** of Tube-A to LEO gas inlet port with icon **IN** (1 in Figure 99)
- 2. Put the cap on LEO gas inlet port with icon = (2 in Figure 99)
- 3. Connect the end the Calibration Kit to **red connector** of Tube-A (3 in Figure 99)



Figure 98. Calibration Kit connected to Tube-A.

- 4. Connect end of the Calibration Kit to the gas cylinder (4 in Figure 99).
- 5. Okolab recommends the use of a purge tube attached to LEO gas outlet port (5 in Figure 99). The purge tube has to be placed in a well ventilated environment. You can use the supplied additional tube as tube for purging.
- 6. Set 1.0 barg (14.5 psig) on the pressure regulator of the gas cylinder (6 in Figure 99).

Note ► Do not exceed 1.5 barg (22 psig).

Tip \blacktriangleright Use the fitting tubes and additional tubes in order to allow the compatibility of the supplied tubing with the gas port of your gas cylinders.



Figure 99. How to connect LEO to N2 cylinder with pressure regulator.

If the N2 cylinder is equipped with a flow regulator, Tube-A and Tube-B are required. Follow the instructions below to connect LEO to N2 cylinder:

- 1. Connect the clear connector of Tube-A to LEO gas inlet port labeled with icon **IN** (1 in Figure 100).
 - 2. Put the cap on LEO gas inlet port labeled with icon (2 in Figure 100).
 - 3. Connect the **red connector** of the Tube-A to the **green connector** of Tube-B (3 in Figure 100)
 - 4. Connect the **end of Tube-B** to gas cylinder regulator (4 in Figure 100).

Tip \blacktriangleright Use the fitting tubes (see paragraph 5.5) in order to allow the compatibility of the supplied tubing with the gas port of your gas cylinders.

- 5. Okolab recommends the use of a purge tube attached to LEO gas outlet port (5 in Figure 100). The purge tube has to be placed in a well ventilated environment. You can use the supplied additional tube as tube for purging.
- Open the gas cylinder regulator by using the valve (6 in Figure 100). Set the flow rate at maximum 0.4 l/min.



Figure 100. How to connect LEO to N2 cylinder with flow regulator.

After connecting LEO to N2 cylinder, press *Ok* (see Figure 97 b). A pop-up with the procedure progress appears, see Figure 97 c.

At the end of the *CO2 Zero Reset* procedure with N2, a pop-up message is shown as in Figure 101. Press *Ok* to return to the *CO2/O2 Calibration* page.



Figure 101. CO2 Zero Reset procedure with N2 completed.

If the *CO2 Zero Reset* procedure has failed the message shown in Figure 102 appears. In this case Okolab recommends to repeat the *CO2 Zero Reset* procedure and if it fails again please contact Okolab support at support@oko-lab.com.



Figure 102. CO2 Zero Reset procedure with N2 has failed.

13.5 Offset

LEO allows also calibration of the CO2 and O2 sensors by using an external gas meter (not hereby provided).

To run the CO2/O2 User Calibration, it is necessary to follow the instructions below:

1. Connect the external gas meter to the incubator in order to read the CO2 and O2 compositions inside the incubator (see Figure 103).



Figure 103. External gas meter connection to the incubator.

Once you have connected the external gas meter to the incubator, wait until it steadily reads CO2 and O2 values and make sure that the system remains stable over time and the incubator is closed. When

you are sure that the system is at the steady state and the read values are stable, take note of the CO2 and O2 values.

- Connect LEO to the incubator at the same point in which you have measured by the external gas meter (see paragraph 10 to connect LEO to the incubator) and start an *Auto* or a *Manual* gas measurement. Read the CO2 and O2 concentrations (see paragraph 10.1.3).
- 3. Calculate the CO2/O2 Offset value (*Offset*) as the difference between CO2/O2 concentration read by the external gas meter (*Reference*) and the concentration read by LEO (*LEO Reading*). Note that also negative values are acceptable.
- 4. Insert the CO2/O2 Offset calculated value (*Offset*) in *Offset* box (as shown in Figure 104 a, b).



Figure 104. How to insert CO2/O2 Offset value (a – b).

5. Insert the CO2/O2 concentration read by the external meter (*Reference*) in *Reference* box and then press on *Ok* (see Figure 105 a, b).



Figure 105. How to insert CO2/O2 Reference value (a - b).

Note ► *When you start the CO2/O2 User Calibration procedure, LEO deletes all stored Offset values.*

14 T1 and T2 modules Calibration

LEO is equipped (on request) with one or two factory calibrated temperature modules (T1-MODULE and/or T2 Module). The factory calibration certificate is included in electronic format in the T1 and T2 modules and can be seen in the T1/T2 calibration pages (see paragraph 14.1).

T1 and T2 modules do not require frequent calibrations. However, you can calibrate them if you are uncertain of their accuracy.

To calibrate T1 (or T2) module follow the instructions below:

- 1. Press the *Calibration* icon in *Settings* menu (see Figure 106 a).
 - 2. Press T1 icon (or T2 icon) to access to the *T1 Calibration* page (or *T2 Calibration* page) (see Figure 106 b)

T1 Calibration page (*T2 Calibration* page) contains three subpages (see Figure 106 c):

- View. To view the factory calibration data or verify if there are offsets on the temperature
- Start. To calibrate the temperature sensor using a calibration procedure

Tip ► You can set until to five temperature values as points for the calibration procedure.

- *Factory Reset.* To cancel the last user calibration on T1-MODULE and restore the factory calibration



Figure 106. T1 Calibration

14.1 View Calibration

Press the *View* icon is (see Figure 107 a) to view the factory calibration data and verify if there are offsets on the temperature sensor readings, as shown in Figure 107 b. If no user calibration was made, there is only the T1 (or T2) Factory Calibration subpage, as in the example shown in Figure 107 b.



Figure 107. T1 Calibration: view calibration (a – b)

14.2 Start Multi-Point Calibration

To perform a Temperature Calibration, you need a certified thermometer as reference and a body of water at uniform temperature (possibly a water bath) then follow the instructions below:

- 1. Connect the temperature module (T1-MODULE or T2-MODULE) that you want to calibrate to LEO.
 - 2. Immerse the certified thermometer and the tip of T1-MODULE (or T2-MODULE) in water bath.
 - 3. Set the water bath to the calibration temperature and wait for the temperature stability.
 - 4. Press *Start* icon (see Figure 108 a).
 - 5. Insert the temperature read by the temperature reference in *Certified Temperature*, as shown in Figure 108 b.
 - 6. Press Set (2 in Figure 108 b) to save the calibration point.

Tip \blacktriangleright You can define up to five temperature points, so you may set the water at any 5 different temperatures within the range 15-55°C.

7. Press *End* if no additional calibration point is required, or iterate the procedure by following points 4,
5, 6 and 7 above to introduce up to five temperature calibration points (see Figure 108 c).



Figure 108. T1 Calibration: start multi-point calibration (a – b - c).

8. When you finish the calibration, an overview appears as in Figure 109. Click on *Save* to complete the calibration or *Cancel* to undo.

23. 24.	71% 😽	12:08
Multi Point C	alibration	
STEP 1 Offset: 0.87 Certified Temperature	25.00	
STEP 2 Offset: 0.88 Certified Temperature	: 30.00	
STEP 3 Offset: 0.87 Certified Temperature	e: 37.00	
STEP 4 Offset: 0.00 Certified Temperature	e: 40.00	
STEP 5 Offset: 0.00 Certified Temperature	e: 50.00	
Save	Can	cel

Figure 109. T1 Multi Point Calibration.

14.3 Factory Calibration

Press *Factory Reset* icon **>** to cancel the last user temperature calibration and restore the factory values (see Figure 110 a). If you are sure of the reset, press *OK*, as shown in Figure 110 b.



Figure 110. T1 Calibration: Factory Reset (a – b).

15 Touchscreen Calibration

Keep pressed the ON/OFF button for 3 seconds to start the Touchscreen Calibration (see Figure 111). While holding the ON/OFF button, the pop-up message shown in Figure 112 a appears. Tap the blue calibration dots until the calibration is complete (see Figure 112 b).



Front view

Right view





Figure 112. Start the Touchscreen Calibration (a – b).

16 Cleaning & Maintenance

Cleaning

For a long lifetime and to take full advantage of this product, Okolab recommends cleaning the product periodically. Please, follow the instructions below to clean LEO:

- Use a polishing cloth or dry cloth to wipe off dust and dirt.
- Do not clean LEO during the charging process.
- Liquids should not enter inside LEO.

Maintenance

 Filters should be regularly inspected and changed if damaged or full of water. Two additional hydrophobic filters are supplied with LEO.



LEO should NOT be used for gas measurement without the filter. Okolab recommends changing hydrophobic filters every three months.

- The CO2-O2-MODULE should be factory calibrated every year and verified by the user each month or before of each important measurement. To calibrate CO2-O2-MODULE, it has to be extracted from LEO. To extract the CO2-O2-MODULE, see paragraph 17.
- The Internal Pump has lifetime of 1000 hours of LEO operations. When the Pump reaches 1000 hours of operations, it should be replaced. Read paragraph 18 to replace the internal pump.
- The Soda Lime kit has to be replaced when the Soda Lime filter changes color turning purple.
 Contact Okolab to purchase a new Soda Lime kit
- The T1 and T2 modules should be calibrated every year.

17 CO2-O2-MODULE extraction

The CO2-O2-MODULE is designed for easy extraction from LEO.

The CO2-O2-MODULE has to be extracted from LEO for the replacement or the re-calibration. Follow the instructions below to extract the CO2-O2 MODULE from LEO:

- 1 Loosen the screw by using a hexagonal key (2.5mm), as shown in Figure 113, a. Then lift up and remove the rear cover of LEO (see Figure 113, b).
- 2 Loosen the screws (1 in Figure 113, c and 2 in Figure 113,d) by using a hexagonal key (1.5 mm).
- 3 Disconnect inlet (see Figure 113, e) and outlet tube (see Figure 113, f) from the CO2-O2-MODULE. Then lift up and remove the CO2-O2-MODULE.



Figure 113. CO2-O2-MODULE extraction.

Follow the instructions below to insert the new CO2-O2-MODULE:

- 4 Connect inlet (see Figure 114, a) and outlet tube (see Figure 114, b) to CO2-O2-MODULE.
- 5 Fasten the screws (1 in Figure 114, c and 2 in Figure 114, d) by using a hexagonal key (1.5mm).
- 6 Put the rear cover on LEO and fasten the screw by using a hexagonal key (2.5mm), as shown in Figure 114,f.



Figure 114. CO2-O2-MODULE insertion

18 Pump extraction

The pump is designed for easy extraction from LEO. It has to be replaced when its lifetime reaches the 1000 hours of operation. As a first step, disconnect CO2-O2-MODULE. Follow the instructions below to extract CO2-O2-MODULE:

- 1 Loosen the screw by using a hexagonal key (2.5mm), as shown in Figure 115, a. Then lift up and remove the rear cover of LEO (see Figure 115, b).
- 2 Loosen the screws (1 in Figure 115, c and 2 in Figure 115, d) by using a hexagonal key (1.5 mm).
- 3 Disconnect inlet (see Figure 115, e) and outlet tube (see Figure 115, f) from the CO2-O2-MODULE. Then lift up and remove the CO2-O2-MODULE.



Figure 115. CO2-O2-MODULE extraction before pump replacement.

Follow the instructions below to replace the pump:

- 1 Loosen the screw (as shown in Figure 116, a) by using a hexagonal key (1.5 mm) and lift up the pump cover (see Figure 116, b).
- 2 Disconnect inlet and outlet tube from the pump (see Figure 116, c).

3 Disconnect the electrical connector of the pump (see Figure 116, d). Gently press the clip to release pressure and remove the connector.

Note ► *Remove the connector with extremely care. You can damage the connector and the port. Damage to the port creates the need for additional repairs to LEO.*



Figure 116. Pump extraction

- 4 Replace the pump with new pump ordered from Okolab.
- 5 Reconnect the electrical connector of the new pump (see Figure 117, a).
- 6 Connect the inlet and outlet tubes (see Figure 117, b). Connect LEO tube marked by ♥ (see Figure 117, c) to the inlet port ♥ of the pump (1 in Figure 117, d). Connect LEO tube marked by ▲ (see Figure 117, c) to the outlet port ▲ of the pump (2 in Figure 117, d).
- 7 Place the pump in its slot (see Figure 117, e) and place the pump cover on it (see Figure 117, f and g).
- 8 Fasten the screw by using a hexagonal key (1.5 mm), as shown in Figure 117, h.



Figure 117. Pump replacement

Follow the instructions below to insert CO2-O2-MODULE:

- 1 Connect inlet (see Figure 118, a) and outlet tube (see Figure 118, b) to CO2-O2-MODULE.
- 2 Fasten the screws (1 in Figure 118, c and 2 in Figure 118, d) by using a hexagonal key (1.5mm).
- 3 Put the rear cover on LEO and fasten the screw by using a hexagonal key (2.5mm), as shown in Figure 118,f.



Figure 118. CO2-O2-MODULE insertion after pump replacement

19 Support

To contact one of our engineers please write to support@oko-lab.com or contact us through the live chat in www.oko-lab.com. You can request a remote support session anytime.

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

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Troubleshooting

The table below shows some frequently asked questions. Please contact Okolab if you need assistance.

Effect	Main Causes	Secondary Causes	Solution	
Auto measurement terms for time-out.	Gas flow rate sent to LEO is lower than values suggested in this manual.		Sent to LEO a gas flow rate higher than 60 ml/min in diffusion mode or 130 ml/min in Aspiration Mode.	
Data download does	USB key is not correctly formatted.		Make sure that USB flash drive is FAT32 formatted.	
not start.	not start. USB key is damaged.		Use a new USB key	
The touch screen does not respond properly, or not at all.	The display is not calibrated.		Press and hold the power button for more than 5 seconds to calibrate the touch screen.	
The device does not log data in internal memory.	Internal memory is full (A warning icon appears on the Homepage.)		Erase all memory data; save data before erasing.	
Humidity Alarm	Possible Moisture Trap absence or wrong connections		Please verify that Moisture Trap is properly connected.	
Possible air infiltration High O2 level and low		Wrong Connections	Verify that all the connectors are correctly plugged on LEO and (eventually) on Moisture Trap	
	Possible air		Verify that Tube B is properly connected on incubator, eventually using fittings provided in the Hard case	
	Pump Leakage	Use and external mass flow meter on the output line of LEO, in order to verify if the Pump flow rate chose is correctly elaborated.		
measurement	surement		Leakage test is required and possibly a Pump substitution.	
	Sensing Cell out of calibration	Wrong offset CO2 and O2 value due to a possible incorrect user calibration procedure	Please restore Factory Calibration	
		CO2 drift due to long term of LEO usage	Please proceed with CO2 Zero Reset and Span Calibration	
Pump stops working	Possible hydrophobic filter occlusion		Please verify that filter and inlet/outlet lines are	
r amp stops working	Possible Inlet/Outlet occlusion	clean	clean	

21.1 LEO

Technical Data		
Maximum pressure	1300 mbar (19 psig)	
Operating flow rate	Min 30 ml/min – Max 400 ml/min	
Barometric Pressure sensor Measuring range	500 – 1200 mbar	
Data Storage	300 measurements	
Data Download	Micro USB to USB OTG host adapter	
Visual and audio alarm	Selectable for each sensor module	
Dimensions	95x155x45 mm	
Weight	492 g	
Power Consumption	10 W	
Power Supply	5V DC	
Environmental Conditions		
Temperature range	5°C – 40°C	
Relative Humidity	0 - 95%	
Barometric Pressure	800 – 1200 mbar	
Pump		
Pump type	Micro Diaphragm Pump	
Pump – Flow	From 60 to 260 ml/min	
Pump Life Expectancy	1000 hours	

21.2 CO2-O2-MODULE

CO2 Sensor – Technical Data		
Sensor type	Non Dispersive InfraRed detector (NDIR)	
Measurement Range	0-20%	
Accuracy	\pm (1% of Full Scale +2% Reading) in the range 0-10% ¹ \pm (2% of Full Scale +3% Reading) in the range 11-20% ¹	
Response Time, T ⁹⁰	< 35 s	
02 Sensor – Technical Data		
Sensor type	Fluorescence-based optical sensor	
Measurement Range	0-25%	
Accuracy	± (1% of Full Scale) in the range 0-10% ¹ ± (2% of Full Scale +3% Reading) in the range 11-25% ¹	
Response Time, T ⁹⁰	< 35 s	
¹ Reference Condition: 1000 mbar and 23°C		

21.3 T1-MODULE

Technical Data		
Sensor	K-type Thermocouple. PFA-insulated: Length 2m; OD: 0.13mm	
Range	0-60°C	
Accuracy (from 20 to 50°C)	± 0.1°C	



Figure 119. T1-MODULE-Technical drawing

21.4 T2-MODULE

Technical Data		
Sensor	PT1000 RTD Class A. Probe Length: 150mm; OD: 2.0mm; Cable Length: 1m	
Range	0-60°C	
Accuracy (from 20 to 50°C)	± 0.1°C	



Figure 120. T2-MODULE-Technical drawing

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Manual Revision Table

Revision Number	Addition of changes	Date
01	Redaction	June 2018
02	Review	July 2018
03	New redaction	November 2018
04	New features	May 2019
05	Changes in hardware	September 2019
06	Changes in tubes	October 2019

WARRANTY

Okolab S.r.l. warrants "LEO" 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. 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. Components which wear are not warranted. Okolab S.r.l. neither assumes responsibility for any omissions or errors nor assumes liability for any damage that may results from improper use of its products in accordance with information provided by Okolab S.r.l. Okolab S.r.l. warrants only the parts manufactured by Okolab S.r.l to be 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. be liable for consequential, incidental or special damage.

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