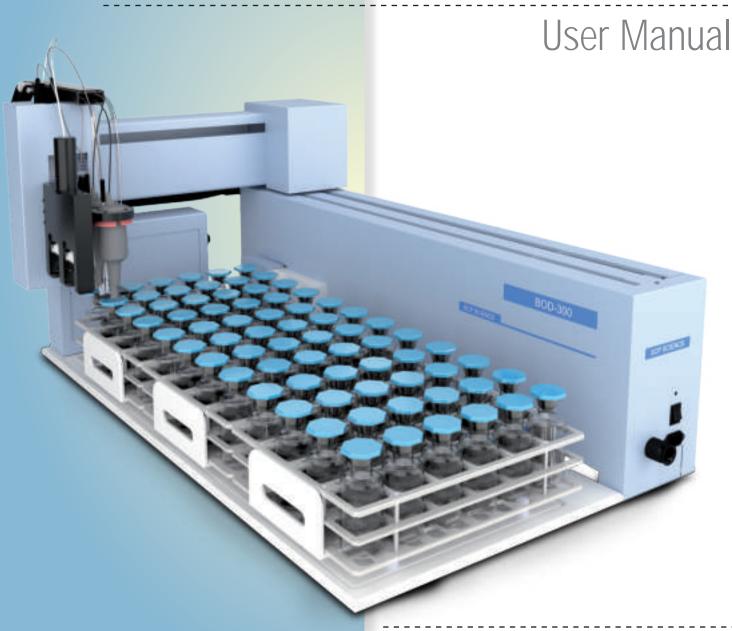


EasyPREP BOD 300



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CORPORATE HEADQUARTERS	USA	FRANCE	GERMANY
21800 Clark Graham Baie D'Urfé (Montréal), Quebec, H9X 4B6 Canada	3 rd Party Distribution Center 348 Route 11, Champlain, N.Y. 12919-4816	12 Ave. de Québec, Bat. Iberis SILIC 642, 91965 Courtaboeuf	Alte Marktoberdorfer Straße 14, 87616 Marktoberdorf
Phone: +1 (514) 457-0701 +1 (800) 361-6820 Fax: +1 (514) 457-4499 +1 (800) 253-5549	Phone: +1 (800) 361-6820 Fax: +1 (800) 253-5549	Phone: +33 (0) 1 69 18 71 17 Fax: +33 (0) 1 60 92 05 67	Phone: +49 (0) 8342-89560-61 Fax: +49 (0) 8342-89560-69

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1 Introduction

EasyPREP BOD-300 is an automated offline system for the sample preparation and analysis for Biochemical Oxygen Demand (BOD). With three liquid-handling peristaltic pumps, the EasyPREP BOD-300 adds seed and nitrification inhibitor as well as dilution water to perform sample dilutions. The low maintenance dissolved oxygen (DO) probe with either optical or membrane technology incorporates a blade stirrer, keeping the sample homogeneous and accelerating probe equilibration for DO measurement. The software manages sample's initial to final DO measurement and BOD calculations. User-definable QC control allows flagging and validation of analytical batches. Preparation and results conform to established methods ISO 5815-1/EN-1899-1/2./5814 (2012), EPA 405.1 and Standard Methods 5210B.

The *EasyPREP BOD-300* accommodates 60 ml, 165 ml or 300 ml bottles for optimized throughput, allowing the analysis of up to 60 position (300 ml bottles), 72 position (60 ml bottles) or 72 position (165 ml bottles) without operator intervention.

Biochemical Oxygen Demand (BOD) analysis is an empirical bioassay-type procedure which determines the dissolved oxygen consumed by microbial life while assimilating and oxidizing the organic matter present in the sample water. The standard test conditions include dark incubation at 20 °C for a specific time period (often 5 days), in an entirely filled and closed bottle. Dissolved oxygen (DO) is measured before and after incubation, and the BOD is computed from the difference.

Carbonaceous biochemical oxygen demand (cBOD) method differs from the BOD method. cBOD includes nitrification inhibitor added in the bottles on Day 0. Nitrification inhibitors available in the market include: allylthiourea (ATU) or 2-chloro-6-(trichloromethyl) pyridine (TCMP). The nitrification inhibitor is added by its own peristaltc pump in liquid form.

Cautions, Notes and Symbols

SYMBOL	DESCRIPTION	SYMBOL	DESCRIPTION
V	Voltage		Mains on
~	Alternating current	0	Mains off
А	Current	<u> </u>	Attention, consult accompanying documents
Hz	Frequency		Protective conductor terminal
F	Fast-acting fuse	W	Watts

Cautions, warnings and notes are included throughout this manual.



CAUTION

A caution is used to emphasize information pertaining to procedures that, if not strictly followed, may result in damage or destruction to the instrument or improper instrument operation.



WARNING

A warning is used to emphasize information about dangerous or hazardous conditions relating to the operation, cleaning or maintenance of the instrument that may result in personal injury.



NOTE

A note is used to emphasize procedures or conditions that may be misinterpreted or overlooked, and to clarify potentially confusing situations.

Warranty & Safety Information

SCP SCIENCE warrants this product free from defects in workmanship and materials for one (1) year from date of purchase.

Should the unit malfunction, please contact SCP SCIENCE's Service Department or your local distributor for further instructions.

The warranty is void if the instrument shows evidence of tampering or has been subjected to excessive moisture, heat, corrosion or other misuse.

SCP SCIENCE shall not be responsible for any damage or losses, however caused, resulting from improper installation or misuse of this product.

Products are supplied for laboratory use only and should not be used for any household, medical or therapeutic application. **SCP SCIENCE** presumes that only trained and qualified individuals, familiar with procedures suitable for the safe operation of these instruments, will handle them. Our customers are solely responsible for the safe operation, handling and use of these products.

MINIMUM SAFETY CONSIDERATIONS MUST BE FOLLOWED WHEN WORKING WITH EasyPREP BOD-300 IN ORDER TO MAINTAIN GOOD LABORATORY PRACTICES:

3.1 MEASURES FOR YOUR PROTECTION



When using chemicals and solvents, comply with the instructions of the manufacturer and the general lab safety rules.



Always wear safety glasses when handling samples and reagents.



No User Serviceable parts inside.

3.2 MEASURES FOR YOUR OPERATIONAL SAFETY



Make sure the EasyPREP BOD-300 is placed on a surface capable of supporting 200 pounds (100 kg).



EasyPREP BOD-300 requires a clearance of five inches (12.7cm) on all sides.



If moving the system, always use 2 people and do not lift by the Kydex® or Corian® panels. Lift by the bottom aluminum frame instead.



Keep hands and fingers free of the system when it is moving to reduce the risk of pinching.



Always plug the EasyPREP BOD-300 in a properly grounded three-prong electrical outlet (100-240V receptacle).



Use a stabilized constant voltage AC power supply, with a voltage within +/-5% of the specified level.



Always use the provided mains power cord or one with the same manufacturer's specifications.



If the **BOD-300** is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.

Speci cations

Dimensions

Width	120 cm (47 in)
Depth (Total)	81.3 cm (32 in)
Depth (Base)	63.5 cm (25 in)
Height	64 cm (25 ¼ in)

Environmental

Relative Humidity	30% to 80%
Altitude	up to 2000 m
Installation	Category II
Pollution	Degree 2
Ambient Operating Temperature	15 °C to 40 °C

Electrical

Model Number	EasyPREP BOD-300
Voltage	115~230V
Power	130W
Frequency	50/60Hz

ProODO Probe Specifications

The specifications listed below are system specifications for the ProOBOD probe when used with a BOD-300 instrument. These specifications are subject to change without notice.

Dissolved Oxygen (mg/L)

Sensor type	Optical (dynamic luminescence quenching lifetime detection)
Range	0 to 50 mg/L
Accuracy	0 to 20 mg/L ± 0.1 mg/L or 1% of reading, whichever is greater 20 to 50 mg/L $\pm 10\%$ of reading
Resolution	0.01 or 0.1 mg/L (auto-scaling)

Temperature (°C)

Range	5 to 70 °C
Accuracy	±0.2 °C
Resolution	0.1 °C

Barometer (mmHg)

zarometer (mmig)		
Range	375 to 825 mmHg	
Accuracy	±1.5 mmHg from 0 to 50 °C	
Resolution	0.1 mmHg	

Fuses-Ratings

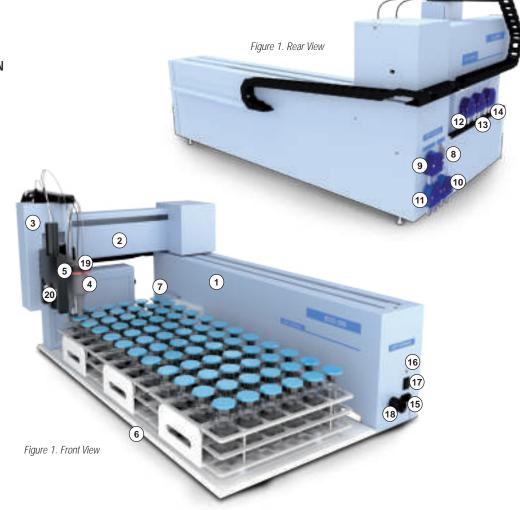


The Power Supply has an over voltage, overload and short circuit protections.

Installation

5.1 BEFORE INSTALLATION

5.1.1 Overview



LABEL	DESCRIPTION
1	X-arm
2	Y-arm
3	Z-arm
4	Dual Probe shown
5	Probe Holder
6	Table
7	Rinse Pump
8	10 pin connector
9	Dilution Pump
10	Seed Pump

LABEL	DESCRIPTION
11	Inhibitor Pump
12	Dilution Pump (Optional)
13	Inhibitor Pump (Optional)
14	Seed Pump (Optional)
15	Power in receptacle
16	Power Indicator
17	Power ON/OFF Button
18	USB Receptacle
19	Autofill sensor
20	Decapper

5.1.2 Unpacking

EasyPREP BOD-300 may be attached to the packaging with bolts. Remove them prior to lifting the system out of the box. The system weighs approximately 100 kg. Use two people to lift by the bottom frame and place the system on a flat surface where it will be used. Save the packing material for further use.

5.1.3 Inspection

Ensure all of the parts are included in the shipment. There may be some accessories that have been packaged in the same box as *EasyPREP* **BOD-300**. If damage is observed, contact the carrier to make a claim and then contact the Service Department.

5.2 HARDWARE INSTALLATION GUIDELINES

BOD-300 requires installation in such a way that the ON/OFF button has a minimum clearance of 8 in. (20 cm)"

5.2.1 Installing The BOD Racks

Place the BOD racks on the top table, using the positioning pins provided on the top table to align them.



Bottle caps must be removed by hand prior to starting sequence, unless the BOD-300 decapping/capping accessory is installed. The BOD-300 decapping/capping accessory is not compatible with acrylic caps. BOD 65 ml, 300 ml PE bottle caps (010-420-020) must be used with the decapping/capping accessory for 300 and 60 ml bottles.

5.2.2 Cable Connections

Make sure that the power button is turned off.

- Connect the power cable to the receptacle located on the end of the X-arm. (Figure 2)
- Connect the USB cable to the USB port located on the end of the X-arm. (Figure 2)



Do not connect the USB cable to the computer yet.



Figure 2. Power and USB ports

5.3 BEFORE SOFTWARE INSTALLATION

5.3.1 Computer System Requirements

PC Processor: 1GHz or superior.

• RAM memory: 1 GB RAM.

• Disk min. space: 1 GB free.

• Ports: 1 USB port available.

• Operating System: Windows 7 and 10.

Software Operation

6.1. OVERVIEW OF SOFTWARE

6.1.1 Software Tabs

The User Interface has four tabs: PROCESS, RESULTS, UTILITIES and ADMIN-TOOLS



Figure 3. Four tabs to navigate the software

- PROCESS tab: Where the sequence is programmed.
- RESULTS tab: Where results are displayed, in real-time.
- UTILITIES tab: Where the basic method parameters can be adjusted, in addition to:
 - 1. Probe calibration and reading.
 - 2. Liquid dispensing parameters.
 - 3. Priming/Cleaning.
- ADMIN-TOOLS tab: Password protected, used to configure the system in the following ways:
 - 1. Motion speed when manually setting the positioning of robotic arm.
 - 2. Liquid dispensing parameters (speed, delay).
 - 3. QC Controls.
 - 4. Method parameters for QC samples

6.1.2 Operational Buttons

There are four (4) buttons on the title bar, as shown below.

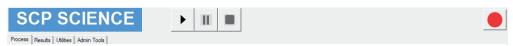


Figure 5. Four buttons to control the test progress

- "Run" button is blue when enabled and grey when disabled.
- "Pause" button is blue when enabled and grey when disabled. It will show as "Resume" after being pressed. When resumed, the sequence will continue from the break point. The pause will only take place once the commands in the que are completed. This may take up to 1 minute to complete.
- "Stop" button goes red when enabled and grey when disabled. When the sequence is interrupted, the test will stop after the current action is finished and the result table is displayed. If the "Run" button is pressed again, the sequence will restart from the beginning.
- "Emergency stop". The system stops completely. The software needs to be closed before restarting the tests.

6.1.3 Hardware Detection



Figure 6. Indicators to show the status of communication with hardware.

The six (6) for dual probe configuration and 4 for single probe configuration indicators at the bottom of the screen indicate whether the modules have been detected by the instrument. A red indicator signifies that a module has not been detected or is faulty. A green indicator signifies that the module has been detected and is ready for use.

ROBOT indicator is for the robotic system.

ProODO #1 indicator is for the first BOD Probe.

ProODO #2 indicator is for the second BOD Probe.

Pump 1 indicator is green to indicate that the software is communicating with the Pump module for the first BOD Probe.

Pump 2 indicator is green to indicate that the software is communicating with the Pump module for the second BOD Probe.

EasyPREP Memory indicator is green to indicate that the software is communication with the on board computer.



If the indicator remains red after start up, go to the Troubleshooting section (Section 10, page 39).

6.2 PROCESS TAB

The PROCESS tab is used to build sequences for automated BOD/cBOD analysis. Sequences can be saved by pressing the save icon or recalled by pressing the search file icon





Figure 7.Process tabs for single and dual probe

6.2.1 Information Field

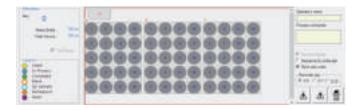


Figure 8. Information field for single and dual probe

Legend: Every line in the sequence is represented by a color in the rack visualization.



Figure 9. Legend field

- Once the "Run" button is pressed, the indicator representing the bottle in process turns blue;
- Once the analysis is complete, the indicator representing the bottle in question turns green.
- If the measurement of the bottle is not within an acceptable for Day 0 reading, the bottle in in question turns orange.
- The system will prompt the user that failed bottles on Day 0 maybe be re-run. The user may remake the bottle, aerate the bottle or accept the value measured. The system will wash the probes between bottles.

Rack: There are 3 possible rack positions: A, B and C.

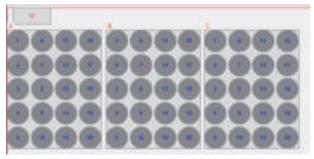


Figure 10. Rack field

- "Operator's Name": A field for the operator's name. This information is stored in the results file. This is only stored after the run button has been pressed.
- "Comment/Remark": A field for any comments. This information is stored in the results file. This information is only stored after the run button has been pressed.



Figure 11. Operator's Name and Comment/Rack fields

6.2.2 Process Sample Field

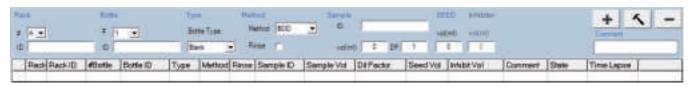


Figure 12. Input sample field

6.2.2.1 Edit sequence tool

- Add task to input process table.
- Makes a correction to selected line in input process table.
- Delete last line or selected line in input process table.
- Clears the entire process table.
- Saves the current task list in the process table.
- Loads a previously saved tasklist.



Figure 13. Edit sequence fileds.

6.2.2.2 Process table fields



Figure 14. Input zone to build test sequences.

- "RACK:" Position of the rack on the rack table. Select between rack positions A, B or C.
- "ID": Unique ID of the rack in use.
- "BOTTLE": Position of the bottle in the rack (range depends on the Rack #).
- "ID": Unique ID of the bottle.
- "TYPE": Select among Blank, Seed, QC, Sample.
- "METHOD": Select between BOD and cBOD.
- "METHOD RINSE": Check to rinse between samples. If rinse is checked the probe is washed between each bottle. Unchecked between bottles and checked between samples to rinse probes between samples.
- "SAMPLE ID": Unique ID of the sample. If the sample ID is the same as another sample the software will calculate the average value based upon it meeting the QC requirements.
- "VOL (ml)": Volume in ml of the sample pipetted into the bottle.
- "DL": Dilution factor. This is the dilution factor prior to aliquotting the sample.
- "SEED VOL (ml)": Volume of seed water to be added to the bottle. Sample and seed volumes will be accepted if the constraint "Sample volume + Seed volume + Inhibitor volume <= Total volume" is satisfied. When the test is running, the first 2/3 of the dilution water is added, then the seed, and then the inhibitor volume request (if a liquid is used). The final 1/3 of the dilution water is added after the seed/inhibitor addition.
- "INHIB VOL (ml)": Volume of inhibitor to be added to the bottle.

6.3 RESULTS TAB

The **RESULTS** tab provides the user with a real-time view of their results. The results are saved automatically into a .csv file located in **BOD-300**/Results folder.



Figure 15. Screenshot of the Results Tab.

Results can be sorted by Probe Pair, by Individual Probe or by Bottle when two probes are used.

- Sorting by Probe Pair will present the results following the analysis order.
- Sorting by Individual probe will list all results from probe 1 followed by probe 2.
- Sorting by Bottle will list the samples in sequential order

6.3.1 Definitions

All the following parameters are stored in the results file.

- "Date": A field for the date of test.
- "Operator": A field for the operator's name.
- "Cell label and cell value": Inspired by an Excel table, in order to show the clicked cell on the results table and its value.
- "Rack #": Position of the rack on the rack table.
- "Rack ID": Unique ID of the rack in use.
- "Bottle #": Position of the bottle in the rack.
- "Bottle ID": Unique ID of the bottle used in the test.
- "Type": Type of sample selected in the sequence and used in the test.

- "Method": BOD or cBOD method selected in the sequence and used in the test.
- "Sample ID": Unique ID of the sample in use.
- "Sample volume": Volume input into the sequence and used in the test.
- "Seed volume": Volume input into the sequence and used in the test.
- "Initial Time": Initial temperature of the sample during Day 0 test.
- "Final Time": Is the time of day sample has been measured.
- "Initial DO": Dissolved Oxygen measured in the initial test.
- "Final DO": Dissolved Oxygen measured in the final test.
- "Final Temp (C)": Temperature of the sample during the final test.
- "Depletion": Calculated by subtraction of final DO from initial DO. .
- "BOD/cBOD (mg/L O₂)": Calculated sample DO depletion.
- "Remark": Comments on data out of range.
- In Remarks, a bottle which failed measuring criteria will be marked with <R>. If remeasured and bottle passed the criteria, <RM>will appear to the right of <R>.

6.4 UTILITIES TAB

In the **UTILITIES** Tab, the following functions are accessible:

- Configuration process.
- Position (Robot).
- · Pump calibration.
- Seed Pump
- Inhibitor Pump
- Dilution Water Pump.
- Waste Pump.
- · Priming/Cleaning.
- ProODO Probe.



Figure 16. Screenshot of the Utilities tab.

6.4.1 Configuration Process

Select sequence test from Day 0, Day 5, Day 7 or Day 20 (figure 18).

- "Day 0": Measurement of the initial dissolved oxygen concentration following the test sequence on day 0.
- "Day 5": Measurement of the final dissolved oxygen concentration on Day 5.
- "Day 7": Measurement of the final dissolved oxygen concentration on Day 7.
- "Day N": Measurement of the final dissolved oxygen concentration on Day N. N is user definable.
- "Select Bottle": Choose capacity of bottles to be used.
- "Total Volume": Actual volume of the BOD bottle.

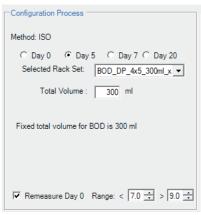


Figure 17. Configuration Process field.

6.4.2 Position (Robot)

- "POSITION (ROBOT)": Manually moves the robot arm to the designated rack and bottle #. When decap is installed the robot will remove caps.
- "Rack": Choose the rack position.
- "Bottle": Choose the bottle position.
- "Use Probe": Move the arm to lower the probe into the designated bottle.
- "Use Pump": Move the arm to align the liquid transfer tubes with the opening of the bottle.
- "Prime All": Used to prime all BOD lines. Robot will move to the wash station.



Figure 18. Position (Robot) field.

)

6.4.3 Calibrate Pumps

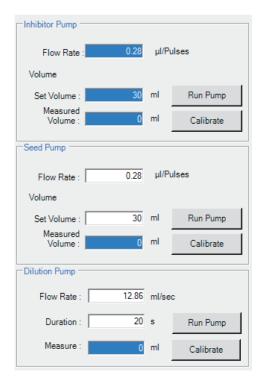


Figure 19. Calibrate pumps.

6.4.3.1 Inhibitor and Seed pumps

Calibration function of the pump for seed/ATU addition (Figure 19).

- Move the robot arm to one of the bottle locations (e.g. A1) in "Position (robot) Use Pump".
- Under "Volume" and "Set": Type in the volume of water to be dispensed by the pump (generally 10-20 ml).
- Press "RUN PUMP" and collect the dispensed water in a pre-weighed bottle.
- Measure the dispensed water, convert to volume and type the value in "Measure".
- Press "CALIBRATE", the new pump flow rate will be reset under "Increment".



Only press Calibrate ONCE.

6.4.3.2 Dilution water pump

Calibration function of the pump for dilution water addition (Figure 19).

- Move the robot arm to one of the bottle locations (e.g. A1) in "Position (robot) Use Pump".
- "Duration": Type in the number of seconds for the pump to run (< 19 seconds).

- Press "RUN PUMP" and collect the dispensed water in a pre-weighed bottle.
- Weigh the dispensed water, convert to volume and type the value in "Measure".
- Press "CALIBRATE", the new pump flow rate will be reset under "Flow Rate".



Only press Calibrate ONCE.

6.4.3.3 Waste pump

Set timing for washing station.

• "FILL": Set the number of seconds to fill the washing/waste station.

6.4.4 Priming/Cleaning

Used to prime the tubing (fill and remove any air bubbles prior to sample analysis) or clean the tubing (fill the tubing with a cleaning solution).

- "INHIBITOR/SEED PUMP": Set volume to prime/clean the inhibitor pump.
- "SEED PUMP": Set volume to prime/clean the Seed Pump
- "DILUTION WATER PUMP": Set volume to prime/clean the dilution water tubing.



Figure 20. Priming and Cleaning

6.4.5 ProODO Probe

- "BAROMETRIC PRESSURE": Reads the barometric pressure.
- "TEMPERATURE": Reads the sample temperature.
- "DISSOLVED OXYGEN": Reads the dissolved oxygen concentration (mg/L 0₂).
- "CALIBRATION": Calibrates the DO probe by one-point calibration in water-saturated air.
- "NEW SENSOR CAP": Input new sensor cap parameters (see section 8.2, page 34).
- "STIRRER": Manually start and stop the probe stirrer.

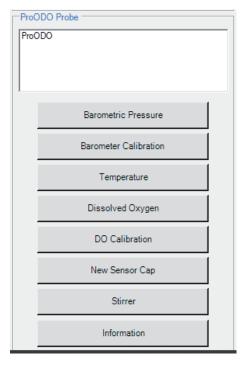


Figure 21. ProODO probe.

6.4.5 Calibration of the BOD probe

Calibrate the DO probe by one-point calibration in water-saturated air. When calibrating, the probe lowers into position A1. The probe should be sealing the opening to the bottle. After reaching 100% water-saturation in the air of the bottle (about 10 minutes), new calibration information is obtained.

- Fill a BOD bottle with 40 ml clean water and place it in position A1.
- Press "Calibrate". When prompted for a calibration time, type "10" and then press Enter.
- New calibration information is obtained and saved.

Though drift is minimal over time, to ensure the best precision, it is recommended to calibrate at the start of each day.

6.5 ADMIN-TOOLS TAB

ADMIN TOOLS is a tab where advanced parameters can be adjusted. This tab password protected (password= scp123)

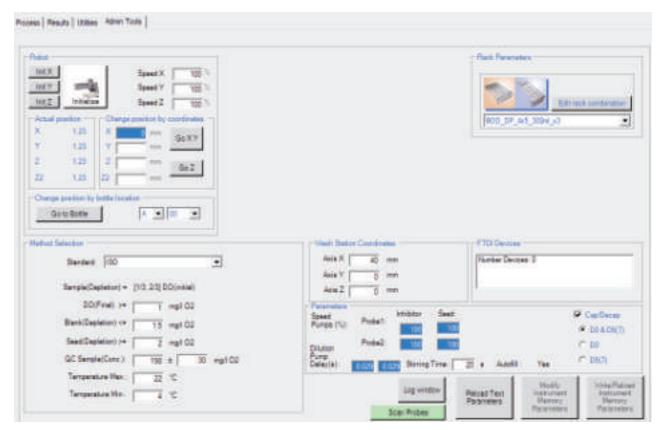


Figure 22. Screenshot of the Admin-Tools tab.

6.5.1 Robot

- "Init X": Individually initializes the X axis. The robotic arm moves the X axis to the home position.
- "Init Y": Individually initializes the Y axis. The robotic arm moves the Y axis to the home position.
- "Init Z": Individually initializes the Z axis. The robotic arm moves the Z axis up to home position.
- "INITIALIZATION": Initializes each axis sequentially starting with Z followed by Y and then X.
- "Axis X": Allow for direct displacement along the X axis, based upon the value given in mm.
- "Axis Y": Allow for direct displacement along the Y axis, based upon the value given in mm.
- "Axis Z": Allow for direct displacement along the Z axis (BOD probe and Autofill sensor), based upon the value given in mm.
- "Axis Z2": Allows for direct displacement along the Z2 axis (Decapper), based on the value given in mm.

- "Speed X": Allow speed adjustment on X axis (1-100-- 1 is the slowest and 100 is the fastest. The default factory setting is 100).
- "Speed Y": Allow speed adjustment on Y axis (1-100-- 1 is the slowest and 100 is the fastest. The default factory setting is 100).
- "Speed Z": Allow speed adjustment on Z axis (1-100-- 1 is the slowest and 100 is the fastest. The default factory setting is 100).
- "GO": Go to selected bottle position, use the drop down menu to select the position. A 00, B 00, and C 00 are coordinates for the wash station.

6.5.2 Rack Parameters

The Rack Parameters section is used to configure and adjust the positioning of the instrument using X, Y and Z parameters. The racks are configured using bottle 1 (back-left corner) of each rack (Figure 23).

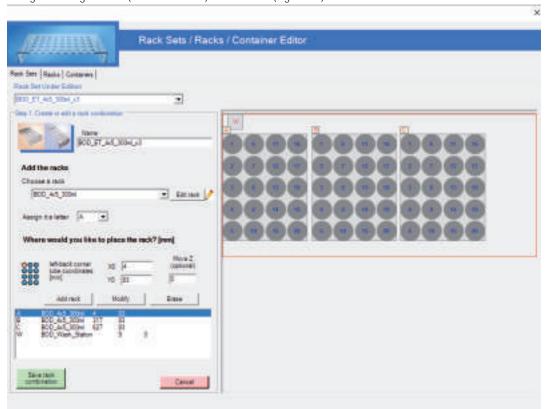


Figure 23. Rack set parameters.

"XO", "YO": Coordinates of the first bottle within the rack.

"MODIFY": Redefines the XO and YO of a highlighted rack using the values placed in XO and YO, respectively.

"CHOOSE A RACK": Allows for different sized bottles to be selected and added to the platform.

"EDIT RACK": Allows for further modification of the racks beyond X and Y positioning.

Once the bottle size is defined as either 300, 165 or 65 ml, the left-back corner tube/bottle is defined by the X0 and the Y0. It is required that the racks parameters are saved once all the required racks have been modified. The wash station parameters can be modified in the Instrument Memory Parameters.

The depth at which the BOD probe enters the bottle to measure the dissolved oxygen is controlled within the racks themselves. "Pressing "Edit Rack" in Figure 23 allow access to further modification of the racks as seen in Figure 24.

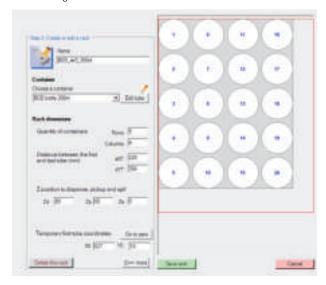


Figure 24. Additional rack modification.

"DXT","DYT": Distance between the first and the last bottle within the rack along each axis. Increase or decrease values of if the probe(s) are not place properly.

- "ZD": Depth of probe when going into the bottle.
- "ZP": Depth of probe when going into the wash station.
- : Allows access to extended settings (decapping height, autofill sensor height, etc....)



Figure 25.

"Z AUTO FILL": Depth of Auto Fill sensor when filling bottles. Increase to reduce fill level and lower to increase the fill level.

"Z CAP/DE-CAP": Depth Decapper Accessory uses to decap bottles

"Y CORRECTION": Offset parameter to account for deviation in height in the Y direction

6.5.3 Calibration of the X, Y and Z positioning for Autofill Sensor(s)

The Autofill sensor can be controlled in the Y and Z by the user. To ensure that the sensor enters the bottle smoothly and is not in contact with the side of the bottle, the X and Y must be calibrated. The Memory can be changed by altering an individual cell and pressing enter. The cell should turn yellow, The Memory parameters must then be updated.

The Memory parameters must then be Written/Reload, which will turn red as it is rewriting. The **BOD-300** must then be restarted.

The X coordinate of the probe is controlled by line 5 in the *EasyPREP* Memory Parameters, as shown in (Figure 26). Alteration of the highlighted value will result in a shift in the x-axis of the Autofill sensor and the pumps. Increasing this value will result a shift to the left and decreasing the value will result in a shift to the right.

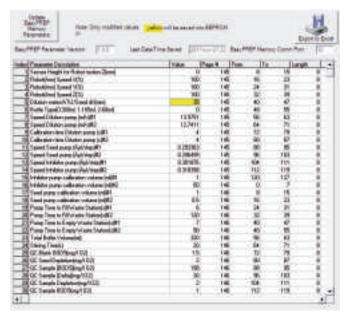


Figure 26.

The Z coordinate of the Auto Fill Sensor is changed in ZAP as seen in (Figure 25). Z auto fill represents the Z value which the Autofill sensor is lowered into the bottle when dispensing liquid. Increasing the Z auto fill will decrease the fill line of the bottles in question and decreasing Z auto fill will increase the fill line of the bottles.

The y- coordinate can be controlled by line 79 in the *EasyPREP* Memory Parameters, and is highlighted below in (Figure 27). This can be a positive or negative value. Alteration of this parameter will bring sensor forward by increasing the parameter and can move the sensor toward the rear by decreasing the parameter.

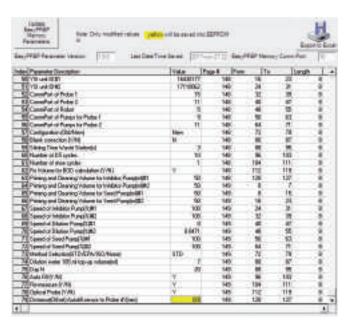


Figure 27.

6.5.4 Instrument Memory Parameters

The Memory can be changed by altering an individual cell and pressing enter. The cell must turn yellow, The Memory parameters must then be updated.

The Memory parameters must then be Written/Reload, which will turn red as it is rewriting. The **BOD-300** software must then be restarted.

6.5.5 QC Controls

None/EPA/ISO: Select input default QC criteria for method selected (EPA or ISO).

"Blank (depletion)": for bottles of Type: Blank, maximum allowed oxygen depletion in the blank. Larger blank depletion values will be flagged as invalid. Default values: EPA: 0.2 mg/L $\rm O_2$ ISO: 1.5 mg/L $\rm O_2$	"Seed (depletion)": for bottles of Type: Seed Control, minimum allowed oxygen depletion in the seed controls. Smaller seed control depletion values will not be used to calculate the seed contribution factor. Default values: EPA: 2 mg/L 0 ₂ ISO: 2 mg/L
"Sample (depletion)": for bottles of Type: Sample, minimum allowed oxygen depletion in the sample. Smaller sample depletion values will be flagged as invalid. Default values: EPA: 2 mg/L $\rm O_2$ ISO formula: (DOI/3) \leq Sample(depletion) \leq (2*DOI/3)	"D0 (Final)": Minimum allowed dissolved oxygen concentration at Day 5/Day7. Lower final D0 values will be flagged as invalid. Default values:
"QC Sample (BOD5)": for bottles of Type: QC Sample. Tolerance range for Glucose-Glutamic Acid standard BOD5 concentrations. Values beyond this range will be flagged as invalid. Default values:	

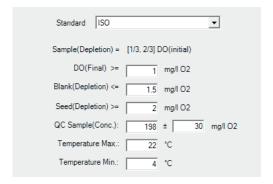


Figure 28. QC parameters.

- "Blank (depletion)": Sets the maximum allowed oxygen depletion in the blank. Blank depletions above this value will be flagged as invalid.
- "Seed (depletion)": Sets the minimum allowed oxygen depletion in the seed controls. Seed controls with depletion values below this will not be used to calculate the seed contribution factor.
- "QC Sample (BOD5)": Sets the tolerance range for standard BOD5 values. Values beyond this range will be flagged as invalid.
- "Sample (depletion)": Sets the minimum allowed oxygen depletion in the sample. Samples with depletion values below this will be flagged.
- "DO (Final)": Sets the minimum allowed dissolved oxygen concentration during Day 5 measurements. Bottles with final DO values below this will be flagged as invalid.
- "Temperature MAX": Max. allowed temperature for samples. Samples above this value will be flagged.
- "Temperature MIN": Min. allowed temperature for samples. Samples below this value will be flagged.
- "Stirring time": Sets the stirring time during DO measurement, and should match equilibration time for the BOD probe.
- If the DayO measurement of the bottle is not within the remeasure range as defined in Utilities, the bottle in in question turns orange. The system will prompt the user that bottles have not passed criteria and will give the user the opportunity to remeasure the bottles in question. The probes will wash between each bottle.
- In Remarks, a bottle which failed measuring criteria will be marked with <R>. If remeasured and bottle passed the criteria, <RM> will appear to the right of < R >.

6.5.6 Parameters

- Seed pump speed: Power setting of the seed pump. Adjusted when installing a new seed pump.
- Timing delay of the dilution water pump. Measured at installation of the pump.
- Stabilization time of the dissolved oxygen probe. Default value: 22 seconds.
- Autofill: Activates or deactivates the autofill sensor
- Cap/Decap: Activates/deactivates the Cap/Decap accessory on each of the runs.



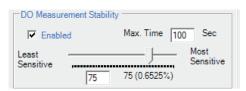


Figure 29. Parameters.

Figure 30. Stability Parameters.

The DO probe use the following criteria to measure a sample. It will take 5 measurements and when the measurements are all with in the sensitivity level the reading is populated on the results page. The factory recommended sensitivity is 95%. The Max. Time is the longest time that a BOD probe will attempt to reach stability. If measurement stability is not reached after this time, the software will note the sample as "not stable".

Guidelines on BOD/CBOD Analysis Using the BOD-300

Before using the instrument for the first time, proceed with the following steps:

7.1 PRIMING

Use the priming function of the software ("Utilities" tab) to fill the dilution water and seed water tubing lines, removing any air bubbles (section 6.4.4).

7.2 PERISTALTIC PUMP CALIBRATION

Verify peristaltic pump calibration of the dilution water and seed/ATU pumps following the procedure described in section 6.4.3 ("UTILITIES" tab).

- Place the dilution water and seed tubing in a container of clean water.
- Use the priming function of the software (in "UTILITIES") to fill the tubing lines with water.

7.3 PROBE CALIBRATION

Perform a calibration of the probe following described procedure (section 6.4.5.1).

7.4 TYPICAL BOD TESTING PROCEDURE

Follow established protocols for the preparation of dilution water, seed, nitrification inhibitor and sample dilutions. Nitrification inhibitor should be added either to the seed water or directly into the sample bottles, manually. In the section below are general guidelines to aid in sample preparation and analysis.

7.4.1 Sample Pretreatment:

- Adjust sample pH if it is not between 6.0 and 8.0 (using dilute H₂SO₄ or NaOH).
- Ensure sample temperature is 20 ± 3 °C.
- A sample's dissolved oxygen level should not be oversaturated. Shake the sample bottle or bubble the sample if needed.

- Remove any residual chlorine (by letting it evaporate or by titration after sodium thiosulfate addition).
- Sample dilution: when diluting the sample, the sample volume should not be less than 1% the BOD bottle volume. Make serial dilutions if necessary

7.4.2 Dilution Water Preparation:

Prepare dilution water according to established methods.

- Use distilled, reverse osmosis, tap or receiving water free of metals and toxic substances.
- Use clean containers free of metals, organics or toxic substances. Preferably use glass containers. Clean containers periodically with a bleach solution.
- Sparge the dilution water with clean tubing dedicated only for this purpose. The DO of the dilution water should be at least 7.5, but not oversaturated. If it is oversaturated, lower the DO level by shaking or sparging the dilution water.

7.4.3 Seed Preparation:

Seed, when needed, as described in established methods.

- Seed source can be from a biological waste treatment system (e.g., settled domestic wastewater, primary effluent, diluted mixed liquor) or from a synthetic seed source. Follow the supplier's instructions on use.
- Seed source should not be filtered but decanted if needed.
- If sparging of the seed water is needed, use clean tubing dedicated only for this purpose.
- If running a cBOD batch, add your nitrification inhibitor to your seed water, or add the inhibitor manually to your BOD bottles.

7.5 QC SAMPLE (GGA STANDARD):

Prepare Glucose - Glutamic Acid (GGA) standard as described in established methods



Note on preparation: Pipette, in each BOD bottle used for QC Samples, an amount of your Glucose-Glutamic Acid standard so that your final concentration will be of 3 mg/L glucose and 3 mg/L glutamic acid. The stock standard preparation being at 150 mg/L, this means a volume of 6 ml of stock standard in a 300 ml bottle, and a volume of 1.2 ml of stock standard in a 60 ml bottle.

7.6 SEED VOL (ml):

The amount of seed should be less when using 60 ml bottles than 300 ml bottles.



Note on preparation: The amount of seed added to Seed Controls should give a dissolved oxygen depletion after incubation of at least $2 \text{ mg/L } O_2$ (see Admin-Tools tab, QC Controls, Seed depletion).

The amount of seed added to QC samples and unknown samples should account for a dissolved oxygen uptake of 0.6 to 1.0 mg/L O_2 , out of the total depletion for that bottle. In addition, the amount of seed should be adjusted from this volume range to that required to provide a GGA result of 198 \pm 30.5 mg/L O_2 for EPA method.

7.7 SAMPLE ANALYSIS

When all the solutions are prepared, turn on the instrument and begin sample analysis.

7.7.1 Day 0

- In "ADMIN-TOOLS": Select one standard method.
- In "UTILITIES": Select Day 0; choose bottle capacity between 300 ml and 60 ml.
- In "PROCESS": Edit test sequence (refer to 6.2.2).



Figure 31. Screenshot of Input sample

The following is an example of BOD analysis sequence.

Rack #	Bottle#	Туре	Method	Sample ID	Sample Volume	Seed Volume
А	01	Blank	BOD	Dilution Water blank	0	0
А	02	Seed Control	BOD	Seed Control #1	0	15
А	03	Seed Control	BOD	Seed Control #1	0	20
А	04	Seed Control	BOD	Seed Control #1	0	25
А	05	QC Sample	BOD	GGA#1	6	5
А	06	QC Sample	BOD	GGA#1	6	5
А	07	QC Sample	BOD	GGA#1	6	5

The other option is to press the "Open" button and select the file 'Demo process' and make the necessary modifications on the populated sequence.

• Click on "Run" button to run the sequence.

Dilution water and seed/nitrification inhibitor are added to the bottles containing samples manually poured in. After liquid dispensing, the probe lowers into the BOD bottle and measures the dissolved oxygen. The probe is rinsed in the rinse station between each sample if rinse is chosen. The Day 0 measurement is reported in column 'Initial DO'.

• Once the analysis is completed, the result table is stored in the folder (path = C:\Program Files (x86)\EasyPREP\Results) and will be needed in the next task.



One can select rinsing between each sample or between different types of samples.

7.7.2 Day 5 or 7

- In "UTILITIES": Switch to Day 5 or Day 7, according to the task specification. Press Ok when warned that the Process list will be cleared.
- In "PROCESS": Select Rack # A. Press "ADD" and select previous run result file to load back the data of tests done on Day 0 from file location C:\Program Files (x86) \BOD300\ ProcD0\ RackA.

The Rack A in the Process Status window will be populated and the Rack A Day 0 data loaded to the Results tab. Same for other racks.

- In Process, select Rack # B. Press "ADD" and select the previous run result file from file location C:\Program Files (x86)\BOD300\ProcDO\
 RackB
- Select Rack # C. Press "ADD" and select the previous run result file from file location C:\Program Files (x86)\BOD300\ProcD0\RackC.
- Place the Racks in their correct position on the BOD-300 platform and uncap all the BOD bottles.



Leaving bottles capped on the platform when the BOD-300 is not equipped with the decapper/capper option can lead to physical damage to the instrument and probe.

• Once all racks result files are loaded, click on the "Run" button to run the process.

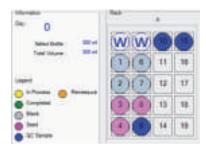
The probe is rinsed at the rinse station between each sample. Sample oxygen depletion and BOD5/7 or CBOD5/7 values are calculated in the "Results" tab, in the columns labelled 'Depletion' and 'BOD/CBOD5/7' respectively.

Once the analysis is completed, the result table is stored in the folder (path = C:\Program Files (x86)\BOD300\Results).

7.7.3 PROCESS TROUBLESHOOTING



Do not pair seed/blank with the QC/sample when using the dual probe BOD-300 such as in Figure 32. The seed efficiency and blank correction must be completed prior to the determination of results. Figure 33 illustrates a proper process.



| Section | Street | W | W | Section | Street | Section | Street | Section |

Figure 32. Incorrect pairing of QC/sample in the process list.

Figure 33. The correct configuration in process list

Ensure the temperature of the rinsing water is 20 °C \pm 2 °C.

7.8 TURNING OFF THE INSTRUMENT

At the end of the run:

- Empty all tubing lines of liquid by using the priming function of the software.
- Periodically, fill the tubing lines with a cleaning solution made of dilute bleach overnight to prevent any biological fouling.
- Remember to keep the sensor and sensor cap of the probe in a humid environment. Follow the sensor storage guidelines in section 8 on probe maintenance.
- Close the BOD-300 software.
- Shut down the computer.
- Power off the instrument.

Guidelines on Probe Operation and Maintenance

8.1 INITIAL SETUP

Remove the ProOBOD cable/probe assembly from the shipping container and locate the instruction sheet that is included. This instruction sheet is important because it contains the calibration coefficients for your sensor cap. After using this sheet for general probe setup, be sure to store it in a safe place in case you need to reload these calibration coefficients in the unlikely event that they are ever deleted from the instrument and probe.



A new cable/probe assembly already has a sensor cap installed and the sensor cap coefficients are preloaded into the probe during production.

Preparing the probe for the first time:

• A new ProOBOD probe will have a clear protective cover placed over the sensor cap to ensure the cap remains moist during shipment. Carefully remove this clear cover from the end of the probe by pulling it straight off the sensor. Save the clear cover along with the sponge for long term storage of the probe. Save the clear cover along with the sponge for storage of the probe.

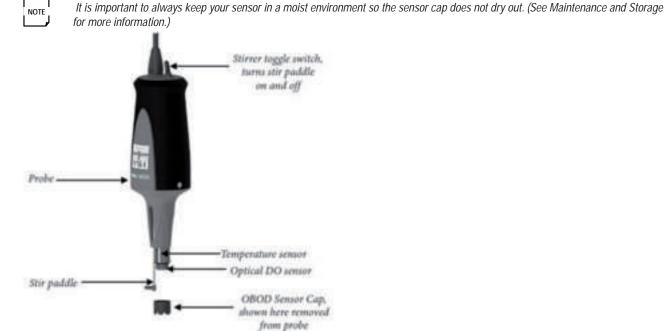


Figure 34. YSI Probe.

8.2 MAINTENACE AND STORAGE

This section describes the proper procedures for care, maintenance and storage of the ProOBOD probe.



Do not attempt to access the probe motor assembly or open the probe body. Doing so will void any remaining warranty. The probe body should only be opened by a YSI Authorized Service Center.

8.2.1 SENSOR MAINTENANCE - DISSOLVED OXYGEN

8.2.1.1 Cleaning the OBOD Sensor Cap

The Sensor Cap should be kept clean since some types of fouling may consume or produce oxygen which could affect the dissolved oxygen measurements. To clean the Sensor Cap, gently wipe away any fouling with a lens cleaning tissue that has been moistened with water. Do not use a coarse towel or cloth or organic solvents to clean the Sensor Cap. Using a coarse towel or an organic solvent to clean the Sensor Cap may cause permanent damage to the cap. for example, alcohol will dissolve the outer paint layer and other organic solvents will likely dissolve the dye in the cap. If the sensor cap is damaged, it must be replaced.

8.2.1.2 OBOD Sensor Cap Replacement

The frequency of replacing the sensor cap is dependent on use. for example, when the probe is powered on for approximately 4 hours per day, 5 days a week, the sensor cap will need to be replaced about once per year. If the probe is powered on more than this, the sensor cap will need to be replaced more often. To extend the sensor cap's usable life, turn the instrument off when not in use. This could mean turning the instrument off over night, but not in between readings. The sensor cap will also need to be replaced if it is damaged or cracked.

When replacing a Sensor Cap, the Sensor Cap Coefficients must be manually updated in the ProODO instrument. The instruction sheet shipped with the replacement OBOD sensor cap includes the calibration coefficients that are specific to your Sensor Cap.



Be sure to save the OBOD Sensor Cap instruction sheet in case you need to re-enter the calibration coefficients.

The replacement OBOD Sensor Cap is shipped in a humidified container and the package should not be opened until the cap is needed. Once the sensor cap has been installed on the OBOD probe as described, it is important to keep the sensor in a 100% humid environment. Therefore, the ProOBOD sensor should be stored either with the clear cap and moist sponge on, in a BOD bottle that contains a small amount of clean water (approximately 40 ml), or in a BOD bottle that is filled with clean water so the sensor cap is immersed in water. See Sensor Storage for more information. If the sensor dries out, refer to the Rehydration procedure in this manual for instructions on how to recondition the sensor cap.

Refer to Figure 35 below when following instructions for replacing the sensor cap.



Figure 35. YSI Probe.



Avoid touching the sensing end of the sensor cap during the following maintenance procedures.

- Remove the stir paddle from the probe by pulling it straight out.
- Remove the old sensor cap from the probe by grasping the probe body with one hand and then rotating the sensor cap counterclockwise
 until it is completely free. Do not use any tools.
- Without the use of tools, carefully remove the O-ring and discard it.
- Clean the O-ring seal on the probe by first wiping off the old O-ring lubricant with a dry lens cleaning tissue. Next, clean any build-up with a water-moistened lens tissue, and then dry with another lens tissue.
- Locate the O-ring supplied in the Sensor Cap replacement kit and install it on the probe. Do not use any tools to replace the O-ring, be careful not to touch the clear optical DO sensor window. After installing the O-ring, ensure it is clean and free of fingerprints. If necessary, wipe clean with a lens tissue.
- Locate the O-ring lubricant included with the new sensor cap. Apply a thin coat of it to the installed O-ring. There should be a thin coat of lubricant on the O-ring only. Remove any excess lubricant from the O-ring and for probe with a lens tissue.
- Clean the clear surface of the optical DO sensor window with a lens cleaning tissue.
- Remove the new sensor cap from its hydrated container and dry the inside cavity of the sensor cap with lens tissue. Make sure that the cavity is completely dry and clean before proceeding with the installation. Using a clockwise motion, thread the new sensor cap onto the probe assembly until it is tight. The 0-ring should be compressed between the sensor cap and the probe. Do not over-tighten the sensor cap and do not use any tools during installation.
- Clean the stir paddle and then re-install it on the probe by pushing it straight into place.
- Store the sensor in a BOD bottle with a small amount of water (approximately 40 ml).
- Follow the procedures below for configuring the ProODO instrument for the new Sensor Cap's coefficients.

8.2.1.3 Configuring the **BOD-300** for the new sensor cap

When receiving and installing a new Sensor Cap, locate the Calibration Code label at the top of the Sensor Cap instruction sheet and note the six codes which are listed as K1to K5 and KC. These six numbers contain the calibration code specific to the sensor cap that was just installed. After installing a new sensor cap, go to the "UTILITIES" tab of the BOD-300 software, press New Sensor Cap. Enter the six numbers of the calibration code, separated by the semicolon ";", then press "Ok".

8.2.1.4 Rehydrating the sensor CAP

The Sensor Cap must remain in a moist environment. If you inadvertently leave your sensor exposed to ambient air for a period of more than approximately 8 hours it may dry out. If the sensor cap is allowed to dry out, it is likely to drift slightly at the beginning of your next study unless it is rehydrated. If the cap dries out, you can rehydrate it by soaking the probe tip with the sensor cap installed in warm (room temperature) tap water for 24 hours. After rehydration, perform a 1-point DO calibration and be sure to store the probe in a moist environment.

8.2.1.5 Sensor maintenance - temperature

You must keep the temperature portion of the sensor free of buildup. Other than that, the sensor requires no maintenance. A soft bristle brush can be used to scrub the temperature sensor if needed. While cleaning, be sure to only scrub the temperature sensor and not the sensor cap.

8.2.2 SENSOR STORAGE

8.2.2.1 Sensor short-term storage

When the ProOBOD is not in use, IT MUST BE STORED IN A MOIST ENVIRONMENT, i.e., the sensor either immersed in water or in water-saturated air. If the sensor cap is allowed to dry out by exposure to dry air, it is likely to drift slightly at the beginning of its next use unless it is rehydrated. If this occurs, follow the rehydrating instructions in this manual. Alternatively, you can place the sensor with sensor cap installed directly in water in a BOD bottle, making sure that the water does not evaporate over time.

8.2.2.2 Sensor long-term storage

For long-term storage (>30 days), remove the batteries from the instrument. Moisten the sponge in the clear protective plastic cap that was originally provided with the probe with clean water, Place the clear cap over the sensor with the sensor cap installed, Inspect the sponge every 30 days to make sure it is still moist, If you no longer have the clear protective cap, place the probe in a BOD bottle that contains a small amount of clean water (approximately 40 ml). Recommended Long-term Storage ambient temperature: -5 to 50 °C (23 to 122 °F).

BOD-300 Peristaltic Pump

9.1 PERISTALTIC PUMP REPLACEMENT

- Empty line from liquid on the pump that needs to be serviced.
- Disconnect inlet and outlet tubing.
- Remove 2 screws that hold the peristaltic pump.
- Replace tube that is inside of the plastic cover.
- Install the 2 screws that holds the peristaltic pump.
- Installation is reverse of removal.
- After installing pump, open the software and verify if the pump is working properly.



Figure 36. Peristaltic Pump.



After servicing peristaltic pump(s), flow rate calibration need to be performed in software for proper accuracy.

9.2 NEW DILUTION PUMP CALIBRATION

- In "ADMIN-TOOLS"/"PARAMETERS", set the "Dilution Water Pump Delay" to 0 s.
- In "UTILITIES", set the "Dilution Water Pump Flow Rate" to 14.5 ml/s.
- Put the end of the dilution water tubing into a bottle of clean water.
- In "UTILITIES", Prime the dilution water pump. Make sure the dispensing line is filled with water and has no air bubbles.

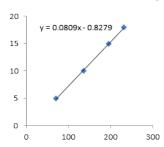
- Weigh 4 empty, clean BOD bottles.
- Dispense water into the 4 empty, tared bottles for 5, 10, 15 and 18 seconds respectively.
- Measure the weight of water in each bottle.

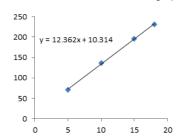
7	Time Delay	0	seconds	
F	low Rate	14.500	ml/s	

Water density applied: 1.00 g/ml. For more accuracy, use the actual density

TIME (s)	EMPTY WT (g)	FULL WT (g)	H20 WT (g)	EXPECTED WT (g)	OFF FROM EXCEPTED (g)	REQUIREMENT
5	26.45	97.21	70.76	72.5	2.4%	<5%
10	26.9	163.03	136.13	145.0	6.1%	<2%
15	26.41	222.22	195.81	217.5	10.0%	<2%
18	26.48	258.42	231.94	261.0	11.1%	<2%

Draw a curve of volume dispensed Vs time and Time Vs volume (see graphs below).





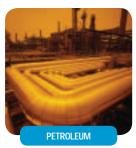
- The intercept (0.827) of the left curve is the Dilution Water Pump Delay (also called Time Delay) of the unit being calibrated.
- Type the Time Delay in the field Dilution Water Pump Delay in the "ADMIN-TOOLS" tab.
- The slope (12.36) of the right curve is the flow rate. Type this flow rate in the field Dilution Water Pump flow rate in the "UTILITIES" tab.
- Validate these settings by repeating step 3-7.
- If the "% off from expected" does not meet the requirements, start again from step 10.
- Once the % off requirements are met, the new dilution pump calibration is finished.

9.3 NEW SEED PUMP CALIBRATION

- Put the end of the seed pump tubing into a bottle of clean water.
- In "UTILITIES", Prime the seed pump. Make sure the dispensing line is filled with water and has no air bubbles.
- Weigh 3 empty, clean BOD bottles.
- Dispense water into three empty, tared bottles for 5, 10 and 20 ml respectively.
- Measure the weight of water in each bottle and convert to volume.
- Calculate the average ratio (ex. 1.055) of volume dispensed/volume required for the three samples.
- In "UTILITIES"/"SEED PUMP", in the field "Set", type the value 1, and in the field "Measure", type the average ratio value (ex. 1.055).
- Press "Calibrate". The new flow rate will be calibrated.

10 Troubleshooting

Topic	Suggestion(s)		
No initialization	- Check if something is blocking one of the arms from moving back to initial position		
Arms not moving	- Switch unit off and check if something is blocking the arms from moving		
Software does not recognize one of the components while starting	- Verify USB cable connection - Verify probe connection		
Inaccurate liquid handling	- Prime fluid lines before using- Check the fittings and tubes (cracks, leakage)- Calibrate the pump		
Pump turning but does not pump liquid	- Check pump tubing and fittings - Replace peristaltic pump tubing		
Invalid DO Value at day 0:	Verify that the proODO probe was properly maintained and not damaged. Assuming that the peristaltic pump calibration, the priming and DO probe calibration were done properly, verify that a well diluted sample pretreatment with a pH 7.0 to 7.2 at 200 °C was used. Verify that the dilution water and the seed were prepared according to the established methods; the QC sample was prepared according to the Glucose-Glutamic Acid (GGA) standard. The dissolved oxygen (DO) in the seed volume is according to the specifications. Then, restart the process. If the same case happens again, contact technical support at SCP SCIENCE .		











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CANADA	USA	FRANCE	GERMANY	UK
Phone: +1 (800) 361-6820	Phone: +1 (800) 361-6820	Phone: +33 (0) 1 69 18 71 17	Phone: +49 (0) 8342-89560-61	Phone: +0 (800) 368-6147
Fax: +1 (800) 253-5549	Fax: +1 (800) 253-5549	Fax: +33 (0) 1 60 92 05 67	Fax: +49 (0) 8342-89560-69	Phone: +44 (0) 7561-7028-90