Quick BOD Meter Model α 1000

Ver. 1.0

Operation & Maintenance Manual





Please read this O&M manual thoroughly and fully understand how to use this analyzer before use.

When using an automatic sample changer, please see the manual for the automatic sample changer.

Caution

The contents of this manual are subject to change without prior notice.

This manual has been created by taking all possible measures. However, if you find any errors or unclear descriptions in this manual, please let manufacturer or the dealer you brought this analyzer know.

Please note that reproduction or copy of part or whole of this manual is strictly prohibited.

Revision History

| Revision | Date | Content |
|----------|----------------|--|
| Rev.1 | May 2009 | Release first revision |
| Rev.2 | June 2009 | revised for addition |
| Rev.3 | September 2010 | AS mode added, tube lengths added |
| Rev.4 | February 2011 | Appendix 3 approximate calculation of reagent |
| | | consumption amount added, the maintenance table |
| | | added at the end of this manual |
| Rev.5 | March 2011 | diameter of wastewater tube changed |
| Rev.6 | March 2011 | additions regarding the air filter at the intake section, as |
| | | well as regarding gas washing container, etc. |
| Rev.7 | March 2012 | Additions regarding aging of trichosporon membrane. |
| | | Additions regarding recycling of trichosporon microbe |
| | | electrode. |



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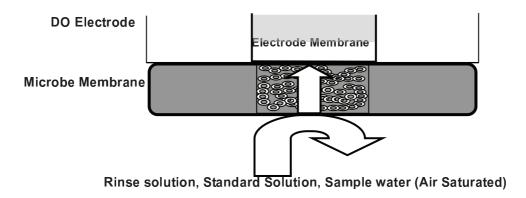
1. Principle of measurement and system outline

Principle of measurement using microbe electrode method (biosensor method)

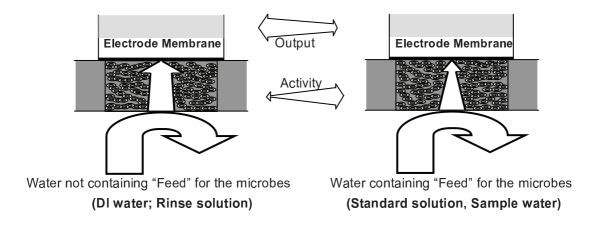
When flushing a solution containing soluble organic matter to a microbe electrode, the microbes in there respire actively and ingest organic matter, resulting oxygen to be consumed. Therefore, the amount of oxygen passing through the immobilized microbe electrode decreases and the electric current to be output from the dissolved oxygen electrode changes. The amount of change corresponds to the concentration of the soluble organic matter. The target to be measured is the components which will become resource in a short time from the soluble organic matter.

When using a microbe electrode in which an immobilized membrane of *Trichosporon cutaneum* (IFO-10466, NBRC-10466) is attached to the sensing section of the oxygen electrode, measuring instruments for biochemical oxygen demands (BODs) using microbe electrode (JIS K 3602) shall be complied to. To avoid misuse of the 5-day method (BOD₅) adopted in the Water pollution control act, BOD to be measured by the instruments complying to JIS K 3602 standard are referred to as "BODs" in this manual.

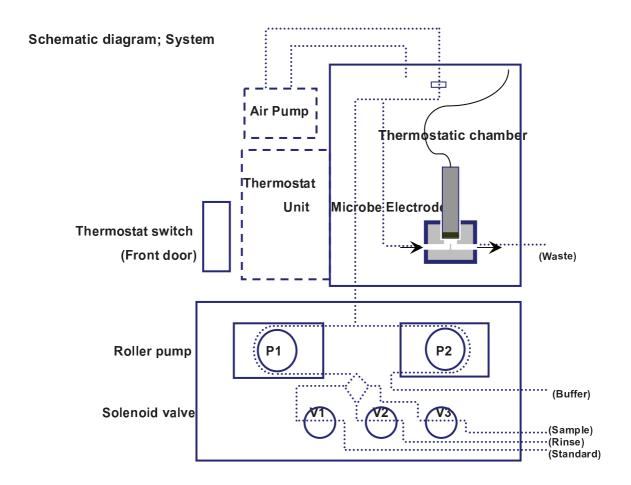
Schematic diagram; Microbe Electrode Tip



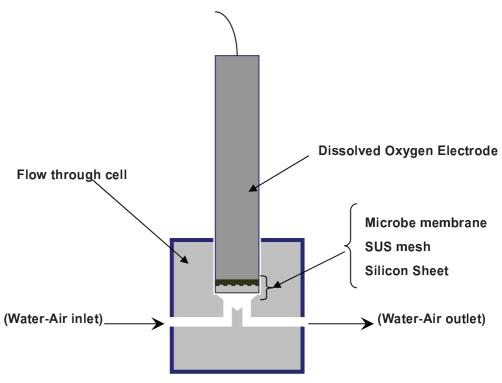
When water containing organic matter ("feed") is provided in a microbe membrane, the microbes in there respire actively and the amount of oxygen passing through the microbe membrane decreases. The decrease of this oxygen amount will be taken as the decrease of the output from the dissolved oxygen electrode.



By comparing the amounts of decrease of the output from the dissolved oxygen electrode measured from both a standard solution and a sample, the sample's soluble BOD concentration is calculated.



Schematic diagram; Microbe Electrode



There is no difference between inlet and outlet.

2. Conditions for installation and precautions for use (For safe use of analyzer)



Before using this analyzer, please read the precautions described in this chapter. In addition, please read this manual thoroughly and fully understand how to use this analyzer before use. Using this analyzer without following to the procedures described in this manual voids the warranty of this analyzer.



(1) Power supply

Be sure to ground (earth) this analyzer. Grounding is also important to ensure analyzer performance by reducing the effects from external noises. As the accessory power cable has a 3-pin connector with grounding terminal, supply power from a 3-pin outlet with grounding terminal. When supplying power from a 2-pin outlet, use the grounding (earth) terminal provided on the main body of this analyzer to ground. Use the power supply indicated in the specifications. At this time, connect to the power supply (current) that satisfy the requirements for stable power supply. In addition, never put too many plugs in one outlet.

(2) Installation environment

Select an appropriate environment (location) for installation and use, satisfying the followings:

Level location

Location with less vibration

Location without significant temperature change

(Do not place the analyzer near the air conditioning or at where subject to air conditioning.)

Location not subject to strong magnetic lines or high frequencies

Location not subject to direct sun light

Location not subject to steam or liquid such as water

Location where ingredients affecting the aspiration of microbes are not contained in the ambient air

Other locations that satisfy the specifications described in this manual.

Particularly, <u>please avoid the following conditions to occur</u> while the analyzer is operating or stopped:

The ambient temperature is lower than 5°C or higher than 30°C.

Leaving the analyzer without operating it with the microbe electrode connected.

^(

(3) Installation stand

Use an installation stand strong enough to support the whole weight when installing the analyzer (approximately 16 kg), reagent solutions, and other equipment together in there.

(4) Roller tube pumps

When a roller tube pump is operating, do not touch its rotating section with your finger or other objects. Be careful not to pinch your finger when connecting/disconnecting tubes.



(5) Valves

When opening/closing a valve, only insert a tube in the open/close section and do not insert other objects such as your finger. Be careful not to pinch your finger when connecting/disconnecting tubes.



(6) Tubing

Make sure that there is no dirt or stains inside the entire tubing system including the flow-through cell before use. Use dedicated tubes for tubing and cut them to the specified lengths to connect. If you find any dirt, stains or scratches on a tube, immediately replace it with a new one. Before performing measurements, ensure that all tubes are filled with a reagent solution.



(7) Reagent solutions

Take care for the main body, parts and connectors not to spill buffer solution, rinse solution, standard solution, sample and other liquids to them. If the main body gets wet, immediately stop measurement and cut the power, and then wipe the spilled liquid and leave the main body stopped until it dries fully.

Please use the buffer solutions and standard solutions available from our company.

Always use new reagent solutions.

Never drink buffer solution and standard solution. Also prevent these solutions from getting into one's eye or putting in one's mouth.



(8) Others

Never disassemble this analyzer and its accessories. Because precise electronic parts and power related parts are mounted inside, touching them could affect human body. Touching them could also cause problem in the analyzer.

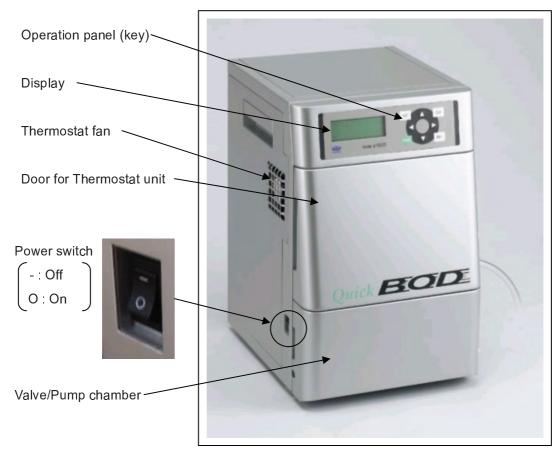
3. List of accessories

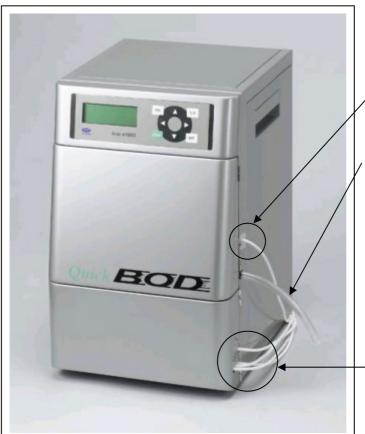
| Item Name | Description | Qty | Storage in |
|------------------------------|--|-----|-------------------------------------|
| Quick BOD α1000 | | 1 | |
| AC Power cord | 3Pins, 10A 125V Included 3Pins – 2Pins converter plus | 1 | |
| Tube set for Quick BOD α1000 | Preset in instrument before delivery | 1 | |
| Air filter | ADVANTEC DISMIC-25CS045AN Preset in instrument before delivery (1) | 6 | |
| Dissolved Oxygen electrode | Polarography DO electrode Including Knurling, Bush, O-ring (contact to flow through cell) | 1 | |
| Flow through cell kit | Flow through cell, Silicon Sheet, SUS mesh | 1 | |
| Membrane kit | With FEP guide ring, 2mil, 5sheet O-ring(1) | 1 | |
| Electrolyte | Electrolyte for DO electrode, 50mL, 1M KCI | 1 | Room temperature |
| Syringe | 2.5mL, for filling electrolyte | 1 | |
| Abrasive | For polishing anode and cathode of the electrode | 1 | |
| Microbe Immobilization set | For ten (10) microbe membrane Porous membrane sheet φ18 (20) abrasive sheet (10) Spacer(10) | 1 | Avoid high temperature and humidity |
| Tools for Immobilization | Stage, Shriner | 1 | |
| tweezers | For microbe membrane | 1 | |
| Bellows pipette | 1mL For preparing microbe membrane | 1 | |
| Plastic container | 100mL, White | 2 | |
| PET vial | PET screw vial, 30mL, colorless | 1 | |
| Resistance | Approx. $1M\Omega$ (equal to $0.6\mu A$ of electrode output) | 1 | |

| BOD SEED | Seed of microbe for measuring BOD, 5 capsule (in plastic case) | 1 | Keeping at 5 -15 degree-C |
|-------------------------------------|---|---|---------------------------|
| Case for accessories | | | (Refrigerate) |
| Printer | Printy2 SD1-31 With printer paper (1) BL-80-30 | 1 | |
| Printer connection cable | RC232C straight D-sub25pin male - D-sub9pin female | 1 | |
| AC/DC adapter for printer | BL-100W INPUT: 100-240V, 100VA, 50/60HZ, 1.0A MAX OUTPUT: 7.2VDC, 5.5A | 1 | |
| AC power cord for printer | | 1 | |
| Phosphoric acid buffer solution | Conc.; 0.5M, Volume; 5L | 1 | Keeping at room temp. |
| BOD Standard | Conc.; 5000mg/L, Volume; 500mL | 1 | Keeping at refrigerate |
| Container for Standard solution | Plastic container, 500mL, White | 2 | |
| Container for Rinse solution | Plastic container, 10L, White | 1 | |
| Container for Buffer solution | Plastic container, 10L, White | 1 | |
| Container for waste water | Plastic container, 20L, White | 1 | |
| Container for drain from thermostat | PP container, 500mL, Colorless | 1 | |
| Washing Brush | H-4 | 1 | |
| O&M manual for Quick BOD α1000 | | 1 | |

The contents of accessories are subject to change without prior notice.

4. Analyzer appearance







Waste (drain) Tube

Drain tube from thermostat unit

Tube for **BUFFER**

Tube for **SAMPLE**

Tube for **RINSE** solution

Tube for Standard solution





Door hinge Upper/Lower door (removal)



Switch for thermostat



Tubing for thermostat

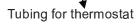


Roller pump

Left: P1 (for Standard and Rinse solution, Sample water)

Right: **P2** (for buffer solution)

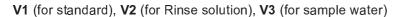
PharMed tube (Yellow) with $\underline{\text{grease}}$





Tubing for valve/pump chamber

Solenoid valve





Terminal for electrode connection

Connect resistance (Approx. $1M\Omega$; equal to $0.6\mu A$ of electrode output) when the analyzer is needed to check electrode output.

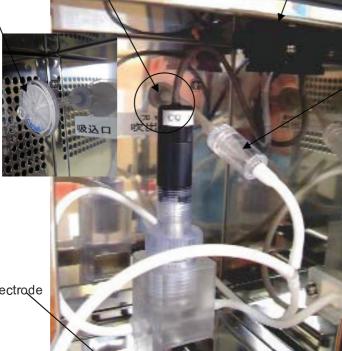


Joint for air pump

Left: inlet (connect air filter)

Right: outlet (connect tube)

(Internal of thermostat chamber)



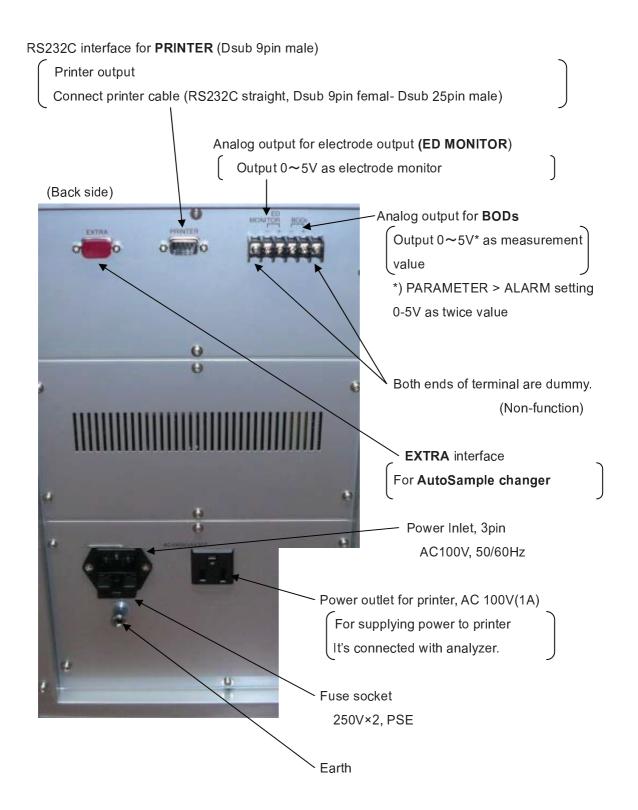
Clack valve (Take care for direction)

Support for electrode

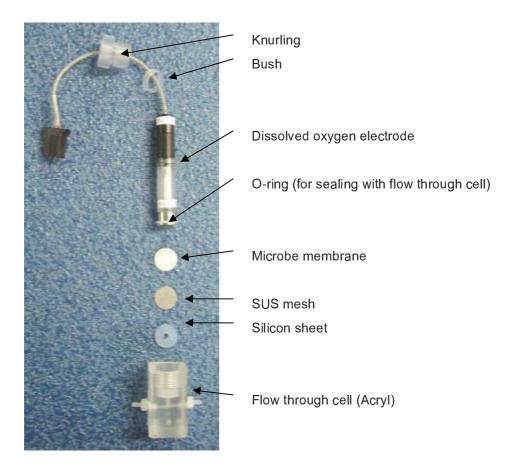


Drain tray

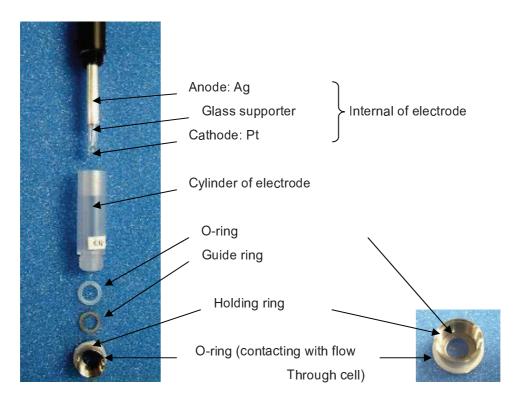
(Connect tube if needed.)



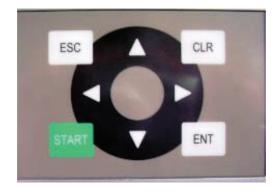
Microbe Electrode



DO electrode (under maintenance)



5. Operation panel/key functions



- ...The start key. Use this key when entering into the measurement mode or other mode from the initial screen. In this manual, this key is indicated as [START] hereinafter.
 - ...Called as the left-arrow key. Use this key such when moving the cursor '__' to the left. In this manual, this key is indicated as [¶] hereinafter.
 - ...Called as the right-arrow key. Use this key such when moving the cursor '__' to the right. In this manual, this key is indicated as [▶] hereinafter.
 - ...Called as the up-arrow key. Use this key to increase a number or when displaying the previous item. In this manual, this key is indicated as [A] hereinafter.
- ...Called as the down-arrow key. Use this key to decrease a number or when displaying the next item. In this manual, this key is indicated as [▼] hereinafter.
- ...Called as the enter key. Use this key to select and run a menu or make setting effective. In this manual, this key is indicated as [ENT] hereinafter.
- ...Called as the clear key. Use this key, such when stopping measurement, together with the [ESC] key. In this manual, this key is indicated as [CLR] hereinafter.
 - ...Called as the escape key. Use this key when changing to the setting mode from the initial screen or when returning to a higher item. In this manual, this key is indicated as [ESC] hereinafter.

In this manual, the left-arrow key and the right-arrow key are collectively indicated as the left and right arrow keys, the up-arrow key and the down-arrow key are collectively indicated as the up and down arrow keys, and all four arrow keys are collectively and simply indicated as the arrow keys.

The indications such as "[ESC] + [CLR]" mean an action to press and hold the [ESC] key first, and then press [CLR].

6. Confirm / Change a setting value

6.1 Content of setting menu

After switching on the analyzer, software version is displayed on screen for a few second, and then default menu is displayed. Press [ESC] key at default screen, setting mode is displayed.









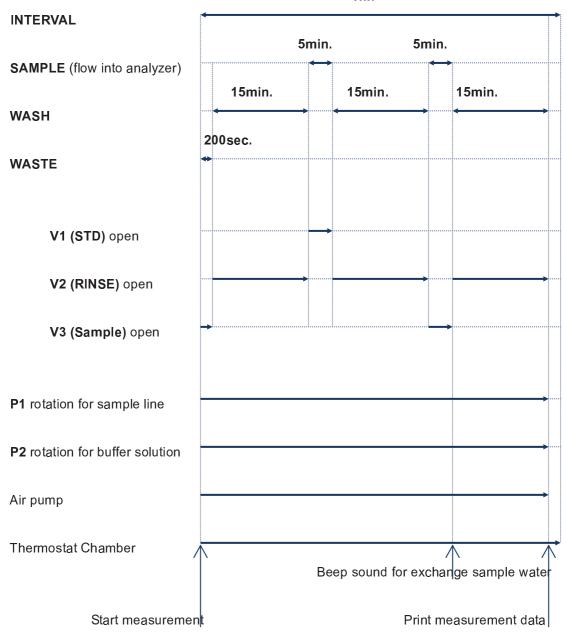
| Main menu | Menu display | Description | Default / Selectable range |
|---------------|--------------|-------------------------------------|---------------------------------|
| 1 PARAMETER | STD | Conc. of standard solution | 050.0mg/L / 000.1-999.9 |
| | ALARM | Setting output span | 050.0mg/L / 000.1-999.9 |
| | SAMPLE | Setting sampling time | 5 min / 060 |
| | WASH | Setting wash time | 15 min / 00-60 |
| | WASTE | Setting drain time | 200 sec / 000-600 |
| | P1 | Select rotation number for P1* | 2 / 1, 2, 3, 4 |
| | P2 | Select rotation number for P2* | 2 / 1, 2, 3, 4 |
| | RATIO ERR | Setting limit of oxygen consumption | 00.0% / 00.0-99.9 |
| 2 SCHEDULE | INTERVAL | Measurement interval | 01:00 hour / 00:00-24:00 |
| | REPEAT | Measurement frequency | 00 / 00 (continuous)-99 |
| 3 SETUP | VALVE | Open/Close each valves | V1, V2, V3 |
| | WARMUP | Warm-up operation | |
| | AUTO FILL | Filling to all tube | |
| | FILL | Filling to each tube | BUF, L1, L2, L3 |
| 4 PRINT | CONDITION | Print setting condition | |
| | DATA | Print measurement data | |
| 5 OPTION | MODE | Select measurement mode | SD / SD, MN, CT, AS |
| | CAL | Select calibration mode | ALWAYS / ALWAYS, ONCE |
| | CHART | Print analog chart of electrode | OFF / ON, OFF |
| | TEMP | Select thermostat temp. | 35.0℃ / 20.0-40.0 |
| | BEEPSOUND | Select beep sound | ON / ON, OFF |
| | SPL BEEP | Select beep at changing a sample | ON / ON, OFF |
| | RANGE | Select scale of electrode output | 2μA / 1, 2 |
| | | Select scale of ED MONITOR | |
| | RESET | Initialization | NO / NO, YES |
| 6 CALENDAR | | Setting Year/Month/Day/Time | |
| 7 EL. MONITOR | | Display electrode output (μA) | |
| 8 SLEEP MODE | SLEEP | Start sleep mode | A-SLP / A-SLP, M-SLP |
| | SLEEP ITV | Sleep mode interval | 03:00hour / 00:15-24:00 |

*) Table for number and flow rate of roller pump P1 and P2

| Selected number | 1 | 2 | 3 | 4 |
|-----------------|------------|------------|------------|------------|
| Rotation | 4rpm | 8rpm | 12rpm | 16rpm |
| Flow rate | 0.5mL/min. | 1.0mL/min. | 1.5mL/min. | 2.0mL/min. |

Measurement Sequence (default setting: SD mode)

1hr.



Measurement mode (5 OPTION > MODE)

| SD mode | Repeat measurement at INTERVAL time | Default setting |
|---------|---------------------------------------|------------------|
| MN mode | Push [START] key for each measurement | Not using normal |
| CT mode | Continues measurement mode | |
| AS mode | Using with auto sample changer | |

Calibration mode (5 OPTION > CAL)

| ALWAYS | Calibrate each sample measurement | Default setting |
|--------|---|------------------|
| ONCE | Calibrate at first time only, then measure sample | Not using normal |
| | water without calibration | |

6.2. STD, ALARM (setting concentration for calibration standard solution, setting span to output measurement value)

Enter conc. for the standard solution to be used for calibration in 1 PARAMETER > STD.

- ① Press [ESC] in the initial screen displayed after turning the power on to change to the setting mode.
- ② Press the arrow keys to move the cursor to 1 PARAMETER, and then press [ENT].
- With the cursor positioned on STD, press [ENT].
- 4 Edit the number with the arrow keys, and then press [ENT].

After the edit is finished, press [ESC] to return to the menu display on higher hierarchy.

The measurement value (Hold) output span (0 - 5V) will be set as twice of the ALARM setting value. Enter concentration to set in 1 PARAMETER > ALARM.

- ① Press [ESC] in the initial screen displayed after turning the power on to change to the setting mode.
- 2 Press the arrow keys to move the cursor to 1 PARAMETER, and then press [ENT].
- ③ Press the arrow keys to move the cursor to ALARM, and then press [ENT].
- 4 Edit the number with the arrow keys, and then press [ENT].

After the edit is finished, press [ESC] to return to the menu display on higher hierarchy.

6.3. REPEAT (setting the measurement count)

Set the number of measurements to be repeated. The set number of measurements completes and the system automatically moves to the sleep operation (when A-SLP setting is selected). Setting the number of measurements to 00 continues measurements repeatedly, and the system no longer moves to the sleep operation. Enter the number of measurements.

- ① Press [ESC] in the initial screen displayed after turning the power on to change to the setting mode.
- Press the arrow keys to move the cursor to 2 SCHEDULE, and then press [ENT].
- ③ Press the arrow keys to move the cursor to REPEAT, and then press [ENT].
- 4 Edit the number with the arrow keys, and then press [ENT].

After the edit is finished, press [ESC] to return to the menu display on higher hierarchy.

6.4. TEMP (setting temperature for the constant temperature room)

Set temperature for the constant temperature room. We recommend setting the temperature in the range of approximately $\pm 5^{\circ}$ C from the ambient temperature. Performing operation in an installation environment where the humidity is high and the temperature is lower than the ambient temperature may cause a formation of condensation in the constant temperature room.

- ① Press [ESC] in the initial screen displayed after turning the power on to change to the setting mode.
- 2 Press the arrow keys to move the cursor to 5 OPTION, and then press [ENT].
- ③ Press the arrow keys to move the cursor to TEMP, and then press [ENT].
- 4 Edit the number with the arrow keys, and then press [ENT].

After the edit is finished, press [ESC] to return to the menu display on higher hierarchy.

Depending on the set temperature, no measurement will start due to that the temperature in the constant temperature room at the time before starting measurement is low (TEMP UNDER ERR). Press [ENT] or [ESC] to return to the initial screen and run warming up

(WARMUP) again in the measurement start operations.

Caution ... If the temperature inside the constant temperature room exceeds 42.0°C, operation stops and a beep sounds. Immediately cut the power.

6.5. SLEEP (sleep mode)

Sleep mode refers to an operation mode to be performed while no operation is performed for the purpose to reduce the consumption amount of reagent solution to be used, comparing to the amount to be used while a continuous measurement is performed. To prevent the sensitivity of the microbe electrode from being degraded, this mode repeats [Standby \rightarrow Rinse solution suction \rightarrow Standard solution suction \rightarrow Rinse solution suction \rightarrow] in a constant interval.

Caution... If using an immobilized microbe membrane, the sensitivity of the microbe electrode may degrade quickly. Please use the sleep mode while considering it.

Setting the sleep mode to automatic start (A-SLP) switches the analyzer to the sleep mode operation automatically after the number of measurements (REPEAT) set beforehand finishes.

The analyzer does not switch to the sleep mode automatically in the following cases:

- In the schedule mode (SD), the number of measurements (REPEAT) is set to 00 (limitless).
- Manual mode (MN) is set.
- In the schedule mode (SD), a measurement is stopped before ending a series of measurements.

The sleep mode operation can be started from the state where measurement is stopped.

- ① In MENU > 8 SLEEP MODE, select manual start (M-SLP) and press [ENT].
- ② Make sure that SLEEP ITV is set to 3 hours, and then press [ENT]. The sleep mode operation starts immediately.

Stopping SLEEP operation

- (1) Press and hold [ESC] + [CLR].
- (2) Press the arrow keys to move the cursor to YES, and then press [ENT].

If an operation is stopped during a process to suction rinse and standard solutions, the analyzer stops after finishing the rinse solution process, and then it returns to the initial screen.

Notes ... For the time setting for the processes to suction rinse and standard solutions in the sleep mode operation, the setting set in MENU > 1.PARAMETER > WASH and SAMPLE will be used. Therefore, the liquid transition time in the interval for one sleep mode operation is (WASH \times 2 + SAMPLE), and the analyzer becomes standby in the remaining time (SW display).

Notes ... The number of sleep mode operations cannot be set.

6.6. Other settings

On routine use of this analyzer, there is no need to change from the initial settings. Follow the above key operations when changing the settings as needed.

7. Preparations for measurement

7.1. Preparation of solutions (rinse solution, standard solution, buffer solution)

Rinse solution

Use clean distilled water.

Storage, use, and limit of quality of standard solution and buffer solution

| | BOD 5000mg/L standard (raw) | 0.5M Phosphate buffer (raw) |
|---------------------|-----------------------------------|-----------------------------------|
| Storage | Keep at refrigerating. | Keep at room temperature |
| | | avoiding high temperature and |
| | | humidity. |
| | | When this is kept at low |
| | | temperature (refrigerate), it may |
| | | crystalize. If crystal appears in |
| | | solution, it needs to keep at |
| | | room temperature or keep at hot |
| | | water for solving crystal. |
| How to use | (Prepare BOD 50mg/L standard) | (Prepare 0.01M buffer solution) |
| | Dilute 100 times exactly using by | Dilute 50 times exactly using by |
| | DI water. | DI water. |
| Expiration date (*1 | 6 month (not opened; stored at | 6 month (not opened; stored at |
| | correct environmental) | correct environmental) |
| Stability (*2 | Use it soon after opened. | Depending on environmental |

^{*1)} Once a solution is opened, the ingredients contained around the air may enter into it or it may be contaminated due to the use of a pipette. Therefore, once opened solutions are out of quality warranty. If you find any abnormality immediately after opened and used a solution, please let our sales department know immediately.

^{*2)} Once a solution is opened, it cannot determine whether its quality limit is exceeded or not, due to that the ingredients contained around the air may enter into it or it may be contaminated due to the use of a pipette. (For determining degradation and contamination, please see the precautions for use.)

Precautions when using standard solution and buffer solution

| | BOD 50mg/L standard | 0.01M Phosphate buffer |
|--------------------------|--|---|
| General notice | Be careful not contaminated from ambient. | Be careful not contaminated from ambient. |
| Tools | Use Mohr pipette or one-mark pipette for preparation of standard. Wash it using detergent and rinse by DI water. Use clean tools. | Use clean tools. |
| Container | Wash container using detergent and brush. Rinse by DI water when preparing standard solution. | Use clean container. |
| Frequency of preparation | Change once per week. *) | |
| Note | Not refilling (mixing) new standard to old standard. Wash the container when filling new standard. | |

^{*)} The higher the ambient temperature, the solution tends to degrade quickly. There is a case that the concentration degraded even in one night, as the container used for the solution was not washed fully.

7.2. Preparation of sludge to be immobilized and points to keep in mind on measurement

Making a microbe membrane using sludge or other substance refers to "Immobilization (of microbe or sludge)", and the created microbe membrane refers to an "Immobilized (microbe or sludge) membrane".

7.2.1. Preparation of sludge to be immobilized

(1) Locations to sample sludge to be immobilized

For the creation of microbe membrane, use sludge (group of conditioned microbes) which is optimum for the sample to be measured. Please select a point to sample by considering the wastewater treatment conditions.

- Example) Sludge near the outlet of the final aeration treatment tank (measurement of treated water)

Although the sludge in the first aeration tank positively responses to the persistent compounds in the flowing water, but it does not tend to response to BOD standard solutions (glucose and glutamine acid solutions).

(2) Pretreatment of sludge to be immobilized

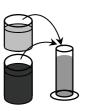
Sludge's internal respiration (cellular respiration) is often active and it continues to consume nutrition in the cell even though it is immobilized. Therefore, there is a possibility that it is difficult to obtain responses against a standard solution or a sample to be measured. Allow microbes to consume nutrition in the cell immediately before immobilizing the sludge. Pour an approximately 10 mL of liquid sludge into a lidded container with a volume of between 50 to 100 mL, and shake it rapidly for several minutes (approximately 5 minutes).

(3) Amount of sludge to be used for immobilization

Use a drop (50 μ L) of sludge (MLSS 5000 mg/L) as a guide.

- If MLSS is thin, collect sludge in a 1L measuring cylinder or similar container and leave it for at least 10 minutes to settle MLSS, and then remove the supernatant liquid. Fully agitate the remaining sludge to suspend them evenly, and then use it for immobilization.
- If MLSS is thick, use smaller amount of sludge or dilute it with a buffer solution (0.01 M phosphoric acid buffer solution) or similar solution, and then use it for immobilization.





(4) When using trichosporon membrane

When using a *trichosporon* membrane, prepare by immersing it in a phosphoric acid buffer solution (diluting a 0.5 M undiluted liquid to 50 to 100 times with distilled water) in room temperature (15 to 25°C) for at least two days and up to one week to one month.

If a dried microbe electrode is assembled and aging operation is started, a longer time will be required to finish aging.

(5) When using BOD SEED

If using BOD SEED, prepare a BOD SEED solution by following to the below procedure: Collect a buffer solution of 10 mL (0.01 M phosphoric acid buffer solution) to be used for measurement into a lidded polyethylene container with a volume of between 50 to 100 mL and add a bran of one BOD SEED capsule, and then shake it rapidly for several minutes (at least approximately 5 minutes).

7.2.2. Points to keep in mind on measurement (determining activity of microbe electrode)

Determine the activity of the microbe electrode according to the data measured from the repeated measurements.

(1) Setting temperature for the constant temperature room

When using a trichosporon membrane or BOD SEED, a temperature of between 32 to 35°C is appropriate.

When using standard activated sludge, a temperature of between 20 to 25°C is appropriate by our experience.

If the temperature of the tank from which the sludge was sampled is high, setting the temperature to that of the tank may be able to well retain the activity of the microbe membrane.

Please set an appropriate temperature based on the activity status of the actual immobilized microbe membrane.

(2) Base level current value output from the microbe electrode

When using a trichosporon membrane or BOD SEED, the base level current value will be stabilized in an operation continued for a half day to several days.

If the amount of sludge used for immobilization was appropriate, the base level current value will be stabilized approximately 1500 to 3500.

If the amount of sludge was large, the base level current value decreases.

If the amount of sludge was small, the base level current value increases. The base level current value shall smaller than the displayed value (D=4095), which is the main body's electrode output threshold range.

If the base level current value becomes large and exceeds D=4095 before fully stabilizing the base level current value and oxygen consumption rate, there is a possibility that the immobilized sludge amount was small. Newly immobilize sludge and create another microbe membrane.

If the base level current value exceeds D=4095 even though an enough amount of sludge was immobilized, there is a possibility that the immobilized sludge was the one that could not retain the activity with only the standard solution and buffer solution. Use the undiluted water to be flew into the sludge tank as the sample (**SAMPLE**) immediately after the start of the aging process (if the standard solution concentration exceeds, use distilled water to dilute).

(3) Responsiveness of the sludge immobilized microbe electrode against BOD standard solution

Even though the microbe membrane immediately after immobilization indicates a response against the sample, on the other hand, there is a case that it does not indicate any response against a BOD standard solution (glucose and glutamine acid solution).

While continuing an operation after assembling the microbe electrode, the response against the BOD standard solution gradually increases and stabilizes. There may be a case that it takes one to several days to be stabilized after starting a continuous operation. Start

sample measurement, at the time when the response against the BOD standard solution becomes large (at least 10% of oxygen consumption rate) and the response stabilizes.

(4) Retaining the sludge immobilized microbe membrane active

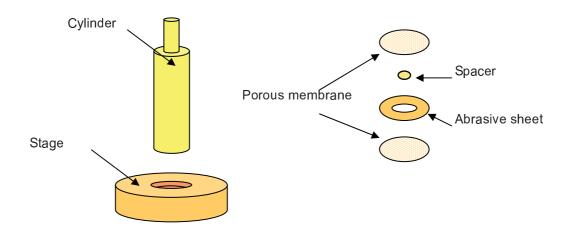
For the purpose to keep the sludge immobilized microbe membrane active, the sample is continually measured during the time other than the measurement time. Use the undiluted water to be flew into the sludge tank as the sample (if the standard solution concentration is exceeded, use distilled water to dilute). Prepare an enough amount of sample. For measuring the sample, a flow rate of approximately 200 mL per 24 hours is required.

Caution ... There is a possibility that the activity of the microbe membrane against the sample will not be retained if only flow water or the treated water having low BOD is measured.

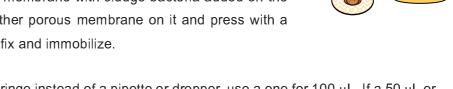
(5) Indications for replacement of microbe membrane

The response against a BOD standard solution may be retained from several weeks to several months. At least 10% of oxygen consumption rate is required. On the other hand, to use a sludge immobilized microbe membrane, the response against a measurement sample decreases gradually from a period, as is often the case. If you find a tendency of which the response against a measurement sample decreases and the BOD measurement values reduces (measurement indication value), immobilize new sludge and create another microbe membrane.

7.3. Procedure to create microbe membrane



- ① Put a porous membrane on the stage. There is no distinction of front and back on porous membrane.
- 2 Peel off one side of abrasion sheet from the two-sided abrasive sheet and put it on the porous membrane. Press it into place with a cylinder tool to adhere enough.
- 3 Take out the assembled sheet and membrane from the stage, peel off the other side of the abrasion sheet from the two-sided abrasive sheet, and put the spacer in the center of the sheet.
- 4 Use a pipette (*1) to put a drop of sludge bacteria (approximately 50 μL) (*2) on the spacer.
- ⑤ Put the porous membrane with sludge bacteria added on the stage. Put another porous membrane on it and press with a cylinder tool to fix and immobilize.



- *1) If using a glass syringe instead of a pipette or dropper, use a one for 100 μL. If a 50 μL or smaller syringe is used, sludge may clog and it no longer suctions it.
- *2) Varies depending on the type or activity of sludge.

Caution ... After creating a microbe membrane, pay attention not to dry it. Promptly assemble it to the microbe electrode and then begin continuous operation.

7.4. Procedure to assemble microbe electrode

Removing the flow-through cell and dissolved oxygen electrode

Open the front door of the main body and disconnect the connector on the upper right portion of the constant temperature room. Remove the tubes connected to the liquid transition joints on both sides of the flow-through cell. Loosen the screws fixing the flow-through cell of the microbe electrode to the flow-through cell support, and remove them together with the microbe electrode as a set. Loosen the knurling screw fixing the dissolved oxygen electrode to the flow-through cell while pushing the screw from above the dissolved oxygen electrode so that the electrode does not turn, and then remove the electrode upward after loosening the screw completely.

Caution ... When removing the connector, push it by pinching both sides of the connector. Do not pull the electrode cable excessively. Otherwise total or partial disconnection may occur inside the cable or the connector.

Caution ... When removing tubes from the flow-through cell, the liquid remaining inside the tubing could spill out of the tip of the tube. To prevent this, disconnect the tube while absorbing the liquid spilled out with a tissue paper or similar material. Do not wet the connector.

Removing used microbe membrane

Using a pair of tweezers, pull up the edge of the used microbe membrane in the flow-through cell to remove it. If the used microbe membrane is adhered on to the tip of the dissolved oxygen electrode, remove it by also using a pair of tweezers.

Dispose of the removed microbe membrane as burnable garbage.

Caution ... Take care so as not to damage the tip (membrane) of the dissolved oxygen electrode with a tip of the tweezers.



Caution ... Take care so as not to damage the microbe membrane, particularly its center portion. Do not disassemble the microbe membrane.

Washing SUS mesh, silicone sheet, flow-through cell

Remove the mesh and sheet from the flow-through cell with a pair of tweezers.

Wash the liquid transition sections of the mesh, sheet and flow-through cell. Use natural detergent or cleanser for washing experimental instruments. Use a narrow brush or similar tool to wash the liquid transition section of the flow-through cell. Fully rinse after washing. If it is very dirty, replace it with a new one. After rinsed, fully wipe off the moisture with a tissue paper or soft cloth.

Caution ... When removing, washing or attaching an SUS mesh and silicone sheet, take care so as not to damage them. Do not bend the mesh, particularly.

Caution ... When washing the flow-through cell, do not use acid solution, organic solution, or alcoholic solution. The flow-through cell could be damaged.

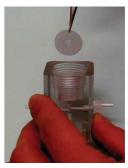
Setting silicone sheet and SUS mesh to flow-through cell

Fully wipe off the moisture from a sheet and mesh and dry them, and then attach the sheet and mesh into the flow-through cell in this order. At this time, never apply excessive force onto them and install them so that they are horizontally attached onto the bottom of the flow-through cell.



Preparing dissolved oxygen electrode

For details on preparing the dissolved oxygen electrode (maintenance work), see the maintenance section. (You do not have to perform maintenance work every time replacing the microbe membrane.)



Attaching new microbe membrane

Never dry out microbe membrane.

Pinch the edge of a microbe membrane and put it in the center of the mesh in the flow-through cell.



Caution ... Take care so as not to damage the microbe membrane.

Notes ... Once a microbe membrane is installed, it is pushed by the tip of the dissolved oxygen electrode, and a slight dent is created at the center of it. Therefore, if reinstalling a once attached membrane, install it in reverse so that the center of the membrane and the tip of the dissolved oxygen electrode (sensing section) are closely attached together.

Installing dissolved oxygen electrode

Before installing the dissolved oxygen electrode, check for moisture on the tip of the dissolved oxygen electrode and wipe it off with a tissue paper or soft cloth if it is wet. Raise the dissolved oxygen electrode vertically and insert it from the top of the flow-through cell until the tip of the dissolved oxygen electrode comes to contact with the microbe membrane horizontally. Hold the tube of the dissolved oxygen electrode as low position of it as possible, and then tighten the knurling screw while pushing the tip of the dissolved oxygen electrode to the microbe membrane. At this time, while taking care so as not to turn the dissolved oxygen electrode, tighten the screw fully until a track of sealing can be confirmed entirely around the O-ring for the flow-through cell.





Caution ... Take care so as not to damage the tip (membrane) of the dissolved oxygen electrode.

Caution ... Take care so as not to offset the microbe membrane position from the center. If it is offset, the oxygen consumption activity of the microbe membrane could not be detected fully at the dissolved oxygen electrode.

Particularly when using a trichosporon membrane, take extra care as its microbe membrane is smaller than the inner diameter of the flow-through cell.

Caution ... Never tighten the screw excessively. Otherwise, the supporting membrane enclosing microbes in the center of the microbe membrane is pushed aside from the center by the protrusion of the tip of the dissolved oxygen electrode (cathode), preventing the dissolved oxygen electrode from detecting the oxygen consumption response of the microbes.

7.5. Setting microbe electrode

After assembling the microbe electrode, put it on the flow-through cell support and tighten the screws for fixing the flow-through cell. When putting the flow-through cell on the support, align the support boss and the fixation hole on the bottom of the flow-through cell and raise the flow-through cell vertically. Connect liquid transition tubes to the liquid transition joints on both sides of the flow-through cell. Connect the connector. Store the electrode cables and tubes in the constant temperature room while avoiding them not to be bent excessively.

Caution ... When closing the front door, make sure that electrode connectors and tubes are not pinched.

8. Aging and measurement

8.1. AUTO FILL

Perform a FILL > AUTO operation at least once to fill fresh solution into the tubing and feed it to the microbe electrode flow-through cell, and then make sure that there is no liquid leakage from the knurling screw fixing the dissolved oxygen electrode, as well as from the liquid transition joints, and other sections. Also confirm the mixing condition of air and solution within the flow-through cell and the waste solution tube. An AUTO FILL operation shall be performed after confirming that all tubes are securely connected.

- ① Press [ESC] in the initial screen displayed after turning the power on to select the setting mode.
- ② Press the arrow keys to move the cursor to menu number 3 indicating SETUP, and then press [ENT].
- ③ Press the arrow keys to move the cursor to AUTO FILL, and then press [ENT].
- 4 AUTO FILL ends approximately 6 minutes and stops automatically with sounding a buzzer. After finished, press [ESC] to return to the menu display on higher hierarchy.

Notes ... If the O-ring for the flow-through cell sealing around the electrode tip is deteriorated, leakage may occur from the knurling screw. If this happens, replace it with a new one.

8.2. Procedure to start aging process (activation)

Continually measure a BOD standard solution of 50 mg/L as a sample. Please prepare this standard solution separately from the container for the standard solution used for calibration. Continuous operations shall normally be continued from one day to several days. If the operation is stopped while aging, the stabilization of the microbe membrane slows significantly, or the microbe membrane may deactivate. Proceed a continuous operation until the oxygen consumption rate increases fully (at least 10%) and stabilizes, and the reproducibility of the measurement values for the standard solution of 50 mg/L falls within ±2.5 mg/L.

Please prepare enough amounts of reagents. As a guide, solution consumption amounts per 24 hours are shown.

| Rinse water (DI water) | 1.1L / 24h | |
|---------------------------------------|-------------|-----------|
| Buffer solution | 1.5L / 24h | |
| Standard solution (as STANDARD) | 120mL / 24h | At aging |
| Standard solution (as SAMPLE) | 200mL / 24h | 320mL/24h |

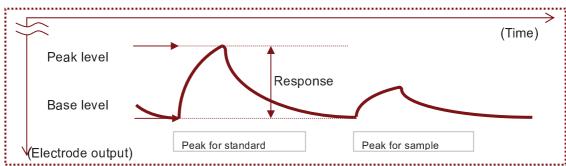
- ① Press [START] in the initial screen.
- ② The system prompts whether to run WARM UP or not. then move the cursor in front of YES and press [ENT]. (Selecting NO immediately starts measurement.)
- ③ Set the time to WARM UP with the arrow keys, and then press [ENT]. WARM UP starts. When WARM UP ends, a measurement begins.

8.3. Measurement data

| 09/04/02 10:12 | → Date/time of starting measurement |
|---------------------------------|---|
| *** START *** | → [Start] |
| MODE =SD | → Measurement mode = (Schedule mode) |
| CAL =ALWAYS | → Calibration mode = (Calibrate always mode) |
| CHART =OFF | → Print electrode output chart = (OFF) |
| TEMP =35.0 °C | → Temperature of thermostat chamber = (35.0 °C) |
| SLEEP =A-SLP | → Sleep mode = (Auto) |
| SLEEPITV =03:00 | → Sleep interval = (3 hour) |
| RANGE =2µAFS | → Electrode output range = (2µA Full scale) |
| BEEPSOUND =ON | → Beep sound = (ON) |
| SPL BEEP =ON | → Beep at changing a sample = (ON) |
| STD =050.0 mg/L | → Concentration of standard solution = (50mg/L) |
| ALARM =050.0 mg/L | → Concentration of alarm (low) = (50mg/L) |
| SAMPLE =05 min | → Sampling time = (5min) |
| WASH =15 min | → Rinse time = (15min) |
| WASTE =200 sec | → Sample wasting time = (200sec) |
| P1 =2 | → Rotation number of P1 = (2) |
| P2 =2 | → Rotation number of P2 = (2) |
| RATIO ERR =00.0 % | → Lower limit of oxygen consumption ratio = (0.0 %) |
| INTERVAL =01:00 | → Measurement interval = (1hour) |
| REPEAT =00 | → Repeat = (00=continues) |
| | |
| ANALYZE=059min | → Time of 1 measurement = (59min) |
| TOTAL=UNLIMITED | → Total = (unlimited) |
| | |
| | |
| 01>09-04-02 10:12 | → Start time of first measurement |
| 51.5mg/l | → Result |
| 20.2% 35.0°C W03:SPL OVER ALARM | → Oxygen consumption / thermostat temp. / Error |
| 0484(0356) 0499(0354) | → Response / Position {Std(Left) / Sample(Right)} |
| 2399-1915 2410-1911 | → Base level / Peak level {Std(Left) / Sample(Right)} |
| | |
| 02>09-04-02 11:12 | → Second measurement |

Note ... When "SKIP" is selected at measurement stop procedure, same parameter of last measurement is printed except start time.

Microbe electrode output chart pattern



Connecting a recorder (input setting: 0 to 5 V) to the electrode output (ED MONITOR) terminal on the back of the main body allows to monitor the patterns like this. If the electrode output is small, change the setting of 5 OPTION > RANGE from 2 μ A to 1 μ A, to expand the chart twice.

In addition, an electrode output chart can be printed using a printer. Set 5 OPTION > CHART to ON beforehand.

What is base level current value (base level)?

A maximum current value during a period from the time when the valve for V1 (for standard solution) or V3 (for sample) opens to the time when the washing process followed to it ends. Indicates the maximum output value of the fully washed microbe electrode.

What is peak level current value (peak level)?

A minimum current value during a period from the time when the valve for V1 (for standard solution) or V3 (for sample) opens to the time when the washing process followed to it ends. Indicates the minimum value of electrode output level decreased due to increase of oxygen consumption activity at the microbe electrode by the water inspection with standard solution or sample.

What is response value?

A value obtained by subtracting a peak level value from a base level current value.

(Response value) = (Base level current value) - (Peak level current value)

Indicates the amount of electrode output decreased (diminution of electrode output) due to increase of oxygen consumption activity at the microbe electrode by the water inspection with standard solution or sample.

What is position?

Indicates the required time [sec] ([second(s)]) to minimize the current value during the period from the time when the valve for V1 (for standard solution) or V3 (for sample) opens to the time when the washing process following to it ends.

What is oxygen consumption rate?

Indicates the magnitude of response of the microbe electrode against a standard solution (that is, the magnitude of calibration curve), by using a ratio between the response value against the standard solution and the base level current value against the standard solution. This value can be obtained from the following formula:

Oxygen consumption rate [%] = (Response value against standard solution / Base level current value against standard solution) x 100

8.4. Procedure to start and continue sample measurement

At the time when the sample suction process finishes in each measurement, a beep sounds to notify that a replacement with another sample to be measured in next time is possible (sample replacement prompt beep). (After the sample collection time ends, the beep sounds 10 times in a 0.5 second interval.) At this time, insert the sample introduction tube into the next sample to be measured. The measurement data will be printed when the washing time ends, following to the sample collection time. According to the measurement interval set in MENU > 2 SCHEDULE > INTERVAL, the next measurement starts.

The sample replacement prompt beep can be disabled if it is not needed. By following to the steps shown below, the setting for 5 OPTION > SPL BEEP can be changed.

- (1) Press [ESC] in the initial screen displayed after turning the power on to change to the setting mode.
- (2) Press the arrow keys to move the cursor to 5 OPTION, and then press [ENT].
- (3) With the cursor positioned on SPL BEEP, press [ENT].
- (4) Set to OFF with the arrow keys, and then press [ENT].

After finished, press [ESC] to return to the menu display on higher hierarchy.

8.5. Procedure to finish measurement

- ① While measuring, press and hold [ESC] + [CLR].
- ② To stop a series of measurement, position the cursor to QUIT and press [ENT]. (To skip a measurement only once, position the cursor to SKIP and press [ENT].)

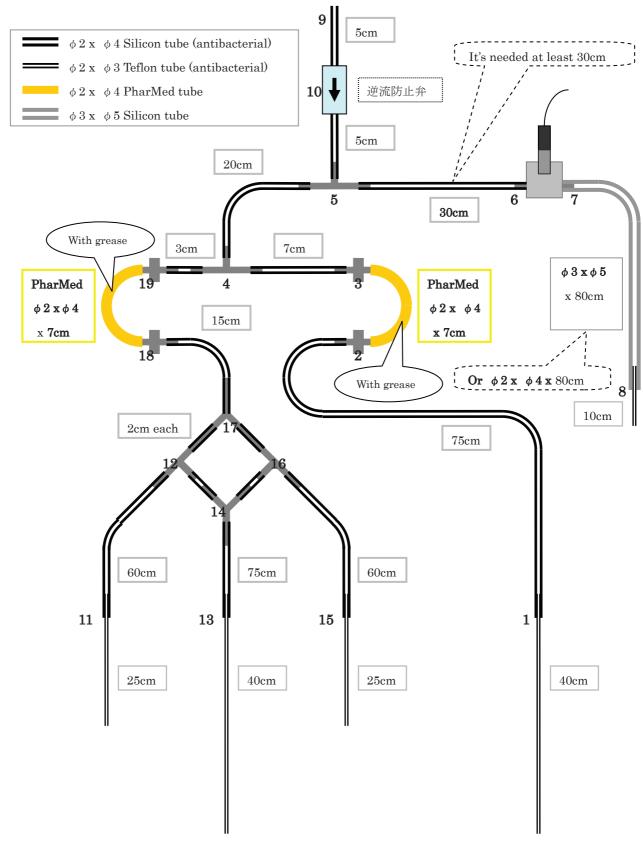
If QUIT (or SKIP) is selected, operations other than the temperature control for the constant temperature room stop after finishing the rinse solution process, and the screen returns to the initial screen as well. (If SKIP is selected, and after the washing process is finished, the next measurement starts in the measurement interval set in INTERVAL, according to the time when the series of measurement is started.)

Caution ... If a sludge immobilized microbe membrane is used, the sample is continually measured in the time other than the measurement time, for the purpose to keep the sludge immobilized microbe membrane active. Do not use the sleep mode. In addition, do not store microbe electrode in refrigerator.

9. Maintenance

9.1. Tubing

9.1.1 Tube diagram



9.1.2. Guide and procedure to replace tube

How slime to be adhered inside the liquid transition line and the rate of slime to be created differ.

Slime found as stain in the liquid transition line is a filmy layer of microbes adhered and grew in the line. Sometimes it can be seen like a viscous mass. If you visually find stains or dirt in teflon tubes, joints, or flow-through cell, immediately replace all the tubes. For the flow-through cell, use detergent and a narrow brush to wash, if necessary.

Sometimes the slime is difficult to find visually. If slime is occurred inside the tubing, the measurement target ingredients will be consumed inside the tubing while transitioning. Use the following procedure to determine the time to replace the tubes (Note that this method might not be used for determination if antibacterial agent or similar material is added in standard solution and buffer solution.):

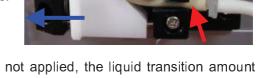
- ① As a sample, measure a standard solution having almost the same concentration of the standard solution that was used for calibration. Please adjust the concentration for a standard solution precisely at this time.
- ② If the result of measurement significantly lower the adjusted concentration for the standard solution, replace the tubes.

Replace the tubes by referring to the section showing the appearance of the main body and the tubing pattern diagram in this manual, as well as the following explanations and pictures.

Installing roller pump tube

For the purpose to prevent your finger from pinching by the roller pump, proceed installation after turning the power to the main body off. Apply grease onto the roller pump tube (φ 2 mm x φ 4 mm, 7 cm PharMed tube).

Insert either of two joints all the way to the bottom of the groove. Pull the upper and lower sections of the lever with your fingers, insert the tube all the way to the bottom so that it runs along the outer periphery of the roller, and insert the other joint into the groove.



Caution ... Be sure to apply grease. If grease is not applied, the liquid transition amount becomes unstable, causing the microbe electrode output unstable.

Inserting into the valve (open/close of each VALVE)

- ① Press [ESC] in the initial screen displayed after turning the power on to select the setting mode.
- ② Press the arrow keys to move the cursor to 3 SETUP, and then press [ENT].
- ③ With the cursor positioned on 31 VALVE, press [ENT].
- 4 Move the cursor to the valve number you want to open,



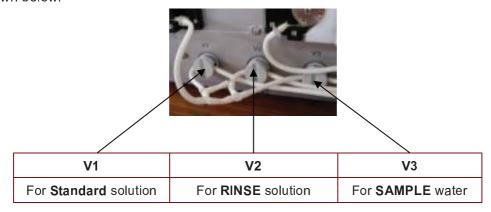


and then press [ENT]. The valves being open are displayed.



(5) With the valve opened, insert a silicone tube from above the valve. When the insertion is finished, press [ENT] and close the valve.

The correlations between each valve and the tube to be pinched shall follow to the table shown below.



After finished, press [ESC] to return to the menu display on higher hierarchy.

Caution ... Do not insert your finger into the valve. Otherwise an accident could occur.

Caution ... Do not insert other than tubes, such as wires or drivers, into the valve. Otherwise a malfunction could occur.

Caution ... Do not leave the valve opened for a longer period without pinching a tube. Doing so may cause the valve difficult to open when reopening the valve from the closed state (this will not happen if a tube is pinched). If this happens, close the valve and leave it for a while, and then try again.

Connecting tube to air outlet

There are two joints on the upper portion of the back of the constant temperature room.

Connect a tube to the air outlet joint on the right side.

For the intake joint on the left side, connect an air filter with a $\phi 2$ x $\phi 4$ tube interposed. (Replace once a month.)



Pinching tube with through-tube section

Pinch a tube all the way to the bottom of each through-tube section.



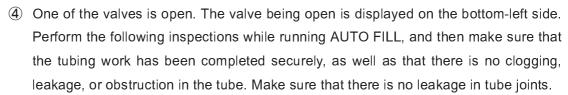




Procedure to fill inside the tubing with solution (running AUTO FILL)

After connecting the microbe electrode and confirming that all tubes are connected securely, fill inside the tubing with solution.

- ① Press [ESC] in the initial screen displayed after turning the power on to select the setting mode.
- ② Press the arrow keys to move the cursor to "3 SETUP", and then press [ENT].
- ③ Press the arrow keys to move the cursor to "33 AUTO FILL", and then press [ENT].



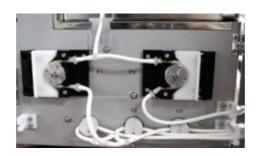
- Confirm that the mixing condition of air and solution within the flow-through cell and the waste solution tube.
- Confirm that a liquid is fed inside the flow-through cell of the electrode unit.
- Make sure that there is no leakage from the knurling screw fixing the dissolved oxygen electrode, liquid transition joints, and other sections.



⑤ AUTO FILL ends approximately 6 minutes and stops automatically with sounding a beep. After finished, press [ESC] to return to the menu display on higher hierarchy. To stop AUTO FILL, press [ESC] + [CLR].

Before closing front door

Make sure that there is no bent or curved section in the entire tubing.





9.2. Dissolved oxygen electrode

9.2.1. Guide and procedure to maintenance of the dissolved oxygen electrode

The longevity of the dissolved oxygen electrode is approximately one year. We recommend to replace it with new one regularly.

As the electrolytic solution in the electrode evaporates in several months, implement maintenance about once per two months.

Removing the external cylinder and tip of the electrode

Hold the dissolved oxygen electrode vertically with the membrane attached section downward, remove the dissolved oxygen electrode and the external cylinder of the electrode by turning them counterclockwise. At this time, take care not to damage the internal pole of the electrode. Also take care for spilling electrolytic solution not to be applied to the connector. Dispose of the remaining electrolytic solution in the external cylinder of the electrode. Turn the external cylinder of the electrode and the membrane retaining screw counterclockwise to remove. Remove the used membrane in the membrane retaining screw by pinching the O-ring (for retaining membrane) in there with a pair of tweezers. Dispose of the old membrane.

Caution ... When removing the O-ring, take care not to damage it.

Washing external cylinder of electrode, O-ring, and membrane retaining screw

There is no need to wash the external cylinder of the electrode, O-ring, and membrane retaining screw. After rinsed with distilled water, fully wipe off the moisture with a tissue paper or soft cloth.

O-rings have a durability limit. If it is pressed and deformed or cracked due to reduced elasticity, replace it with a new one. These criteria are also applied to the O-ring (for close contact of flow-through cell) for the flow-through cell attached to the outer periphery of the membrane retaining screw.

Polishing electrode internal pole

After rinsed with distilled water, remove the moisture fully by absorbing it with a piece of Kimwipe or soft cloth. Take a small amount of abrading agent on another piece of Kimwipe or similar material. Spread it on a flat, smooth surface table.

Apply the electrode tip (cathode) to the abrading agent and polish it. At this time, hold the electrode vertically and move it gently in a circular motion on the piece of Kimwipe or similar material. Next, using another piece of Kimwipe or similar material on which no abrading agent is placed, repeat the same motion (vertically holding the electrode and move it in a circular motion) and remove the abrading agent. The electrode tip (cathode) is made of platinum. After polishing, it shows a shine of platinum.

Using a piece of Kimwipe or similar material with abrading agent taken on it, polish the side of the electrode (anode). Next, using another piece of Kimwipe or similar material on which no abrading agent is placed, repeat the same motion and remove the abrading agent. The side of the electrode (anode) is made of silver. After polishing, it shows a shine of silver. Polish the side of the electrode (anode) so that almost half of its surface shines.

After polishing, rinse it with a plenty amount of clear distilled water.

Caution ... Do not apply excessive force for polishing. Otherwise the internal pole of the electrode may be damaged. In addition, take care not to damage or contaminate the electrode tip (cathode) and the side of the electrode (anode). Pay particular attention to the electrode tip (cathode), because scratches or dirt that cannot be seen by human eye may cause faulty symptoms such as excessive output.

Caution ... If rinsing after polishing was not enough, the electrode output after assembling may become unstable, or may increase residual current (excessive output), which shows the same pattern at the time when the electrode life ends.

Installing knurling screw and bush

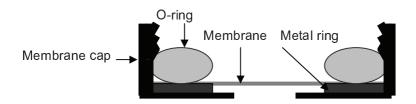
Knurling screw and bush can be removed from the dissolved oxygen electrode through its internal pole, by removing the external cylinder of the electrode (membrane retaining screw) from the dissolved oxygen electrode.

Insert the knurling screw and bush in this order, to where they were removed, before performing the next step. Take care for the direction of the knurling screw.

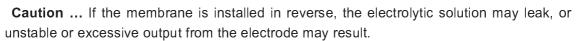
Attaching membrane and O-ring

Insert a new membrane into the membrane retaining screw. A membrane is affixed to a metal ring from its one side. While facing the metal ring side downward (the side for membrane retaining screw) and the membrane affixed side upward (the side to be pressed by an O-ring later), insert the membrane horizontally so that it is in place to the bottom surface of the membrane retaining screw.

After inserting the membrane, insert an O-ring (for retaining membrane). After inserting the membrane into the membrane retaining screw, lightly press the edge of the O-ring with a pair of tweezers, and then insert the membrane horizontally so that it is in place to the bottom of the membrane retaining screw.

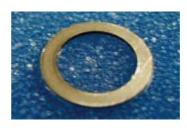


Insert and turn clockwise the external cylinder of the electrode into the membrane retaining screw in which the membrane and the O-ring are inserted horizontally, and then tighten fully.





Upper side



Lower side

Caution ... Be sure to use a new membrane when reassembling the dissolved oxygen electrode. The used membrane might be elongated as it was pressed by the electrode tip (cathode), resulting poor contact with the electrode tip (cathode).

Caution ... When holding a membrane, pinch the metal ring from its outer side so as not to touch the membrane directly (never apply scratches or dirt).

Assembling dissolved oxygen electrode

Face the membrane retaining screw downward and raise and hold the external cylinder of the electrode vertically, and then take electrolytic solution with a syringe and pour it up to a half of the height of the external cylinder of the electrode (approximately 1.8 to 2.0 mL). Hold the upper side



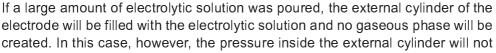


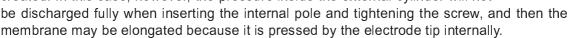
of the external cylinder of the electrode and snap the lower side of it with your finger to float and remove bubbles accumulated in the tip of the external cylinder.

Raise and hold the dissolved oxygen electrode vertically and insert its internal pole gently into the external cylinder of the electrode raised and held vertically and filled with the electrolytic solution. By seeing the external cylinder of the electrode from above, slowly turn and tighten its screw counterclockwise. Tighten the screw completely. If the electrolytic solution spills, rinse it with distilled water, and then fully wipe off the moisture with a tissue paper or soft cloth.

Caution ... Turn slowly when inserting the internal pole and tighten the screw. If it is tightened quickly, the pressure inside the external cylinder could not be discharged, causing the membrane at the electrode tip pushed internally and elongated.

Caution ... When the poured electrolytic solution amount is appropriate, a certain gaseous phase will be created in the top section of the external cylinder of the electrode. As the amount of electrolytic solution to be poured varies depending on individual electrodes, please adjust separately.





Caution ... When pouring electrolytic solution into the external cylinder of the electrode with a syringe, take care so as not to create bubbles in the electrolytic solution. The bubbles remained around the membrane in the electrode cylinder may cause the activity of the dissolved oxygen electrode unstable.

9.2.2. Confirming output from the dissolved oxygen electrode

If necessary, confirm the output from the electrode using a single dissolved oxygen electrode before assembling the microbe electrode.

Notes ... For details on how to confirm the normal output of the dissolved oxygen electrode see the following table. In either case, connect a single dissolved oxygen electrode to the connector of the main body and confirm with the EL.MONITOR display. As this electrode uses polarography, the output cannot be confirmed with standard testers.

| Confirmation | Procedure | |
|-------------------------|--|---------------|
| Insulation failure | Soak 5% Sodium sulfite solution at room | Less than |
| Peeling off of Pt/glass | temperature | 0.1µA, Stable |
| Response (90%) | Move the electrode from air to 5% Sodium | Within 60sec |
| | sulfite solution. Measure time to decreasing | |
| | 90% of first reading. | |

^{*)} Peeling off of electrode tip (cathode) platinum (Pt) section from its supporting glass section

Determining output condition from the dissolved oxygen electrode

| Electrode | Cause | Action | | | | |
|---------------|---|---|--|--|--|--|
| output in air | | | | | | |
| unstable | Dark current or Peeling off of Pt/glass *) | Reassemble the electrode or replace to new electrode. | | | | |
| | Loose connector | Need to replace to new electrode. | | | | |
| | Cable disconnection | | | | | |
| | Air bubble remains in tip of electrode (cathode). | Reassemble the electrode. Tap the side of the electrode cylinder to dislodge any air bubbles left in the electrode. | | | | |
| | Abrasive remains on surface of internal electrode | Remove abrasive using paper tissue and rinse they electrode by DI water. Fill new electrolyte and assemble the electrode. | | | | |
| Too small | Air bubble remains in tip of electrode (cathode). | Reassemble the electrode. Tap the side of the electrode cylinder to dislodge any air bubbles left in the electrode. | | | | |
| | Shortage of electrolyte (or empty) | Reassemble the electrode. | | | | |
| | Polishing of electrode is not enough. | Reassemble the electrode. (Polish surface of internal electrode.) | | | | |
| | Abrasive remains on | Remove abrasive using paper tissue and rinse | | | | |
| | surface of internal electrode | they electrode by DI water. Fill new electrolyte and assemble the electrode. | | | | |
| Too big | Dark current or | Need to replace to new electrode. | | | | |
| | Peeling off of Pt/glass *) | | | | | |
| | Membrane is getting older. | Reassemble the electrode using new membrane. | | | | |
| | Membrane is upside down. | Reassemble the electrode. | | | | |

^{*)} A symptom in which electric resistance between the electrode tip (cathode) and the side of the electrode (anode) reduces due to peeling off of the electrode tip (cathode) platinum (Pt) section from its supporting glass section.

Notes ... When confirming the input function of the main body, separately from the output confirmation of the dissolved oxygen electrode, connect a dummy resister of approximately 1 $M\Omega$ to the electrode connector connection terminal and confirm with the



EL.MONITOR display that the electrode output is approximately 0.6 μA and stable.

10. Troubleshooting

Check the following items which are required for normal operation and reliable performance, and perform appropriate countermeasures such as change of installation location, request for analyzer repair, replacement of reagent or tube, and maintenance of the dissolved oxygen electrode.

| <u></u> | Check the power (AC) |
|--------------------|---|
| environmental | Connect earth. |
| mu | Ambient temperature should be 5 - 30°C. Not changing suddenly. |
| \ \ Si | Not installed at near the air conditioning system. |
| e | Not containing interference material in ambient air. *) |
| | Valves operate correctly. |
| | Need to check from "SETUP>VALVE". |
| | Roller pump operates correctly. |
| Je. | Need to check from "SETUP>WARMUP". |
| Analyzer | Air pump operates correctly. |
| An | Check air blowing from tip of waste tube. |
| | Thermostat chamber operates correctly. |
| | Check panel display temperature (T =) and temperature in measurement data. |
| | Electrode output is correct. |
| | Prepare enough volume of reagents. |
| | No contamination in reagents. |
| Jce | Use clean DI water for rinse solution. |
| enal | Tube is set correctly. |
| Maintenance | Tube does not dirty or no color. |
| ĕ | Tube is not twisted. |
| | There is no back pressure for waste tube. |
| | Not remaining microbe electrode (not remaining internal analyzer). |
| | Reproductively of base level current is stable. There is no big change. |
| tivit | Reproductively of oxygen consumption ratio current is stable. |
| ac | Oxygen consumption ratio is over than 10%. |
| epo. | Measurement result should be less than 2mg/L when measuring same grade water as rinse |
| Electrode activity | solution. |
| | When diluting standard solution to 2 times, measurement result become half of original value. |

^{*)} For example, ethanol contained in the ambient air dissolves quickly into the solution in the tubing through the air pump and it significantly affects the activity of the microbe membrane, regardless of the valve operation sequences.

11. Specification

| Instrument | : | Quick BOD α1000 |
|----------------|---|--|
| Method | : | Bio-Sensor method (Microbe electrode) |
| | | JIS K3602 Apparatus for the estimation of biochemical oxygen demand (BODs) |
| | | with microbial sensor (using <i>Trichosporon cutaneum</i> membrane) |
| Sample water | : | Industrial water, river water including soluble organic matter |
| Range | : | 2~50mg/L |
| Interval | : | 60min / 1 sample |
| Calibration | : | Single point calibration using by BOD standard (Glucose-Glutamic acid) |
| Reproductively | : | ±5% (F.S.) |
| | | (using Trichosporon cutaneum membrane, temperature is stable) |
| Resolution | : | 0.1mg/L |
| Thermostat | : | 20 – 40 °C |
| Output | : | Printer output |
| | | Data output (DC 0 - 5V) |
| | | Electrode output (DC 0 - 5V) |
| Power | : | AC 100V ±10V, 50/60Hz, 3A max |
| Temperature | : | 5 – 30 °C (not changing suddenly) |
| Humidity | : | < 90% RH (non-condensate) |
| Sample temp. | : | 10 °C - (Temperature of thermostat) |
| Dimension | : | 260(W) × 320(D) × 409(H) mm |
| Weight | : | 16 Kg |
| Accessories | : | Microbe Immobilization set (membrane and tools), |
| | | Printer, |
| | | Standard solution, Buffer solution etc. |
| Option | : | Microbe membrane (Trichosporon cutaneum membrane) |
| | | Pipette etc. |
| | | Autosample changer AS1000 (for 11 samples) |

(Appendix 1) Contents to be set for accessory printer

Select parameter of Printy3 BL-80RS II

| Parameter | Setting Parameter with BOD analyzer | Comment | | |
|--------------------|-------------------------------------|-----------------------------|--|--|
| (Data input) | (Serial) | | | |
| International Char | Japan | (Default) | | |
| Print mode | Graphic | (Default) | | |
| Character set | 24Dot ANK Gothic type | (Default) | | |
| Select switch | Invalidity(OFF) | ← Change from Available(ON) | | |
| Baud rate | 9600bps | (Default) | | |
| Bit length | 8 bit | (Default) | | |
| Parity | Non | (Default) | | |
| Data control | SBUSY | | | |
| Paper selection | Normal paper | (Default) | | |
| Uprignt/inverted | Upright printing | (Default) | | |
| Auto Power Off | Invalidity(OFF) | ← Change from Available(ON) | | |
| Graphic speed mode | Low speed | (Default) | | |
| Battery mode | Invalidity(OFF) | | | |

How to confirm or change the settings for the operation functions (See the printer's operation manual.)

Turning the power on while pressing and holding the FEED switch prints the current settings.

If you do not change the settings, press the FEED switch. A test print is implemented and the operation setting mode ends.

If you make a change to the settings, press the SELECT switch. The operation setting mode is selected.

While in the operation setting mode, use the FEED switch and the SELECT switch to confirm or change the settings for each setting item sequentially.

Pressing the FEED switch proceeds to the next setting item and prints the contents of it.

Pressing the SELECT switch changes the setting contents and prints the changed setting contents.

Proceeding confirmation or change of the setting contents in this manner performs a test print finally, and then ends the operation setting mode.

Caution ... Do not connect any device other than the dedicated printer to the 100 V AC (1A) power output terminal for printer on the back of the main body.

(Appendix 2) Content and preparation of reagent

BOD 5000mg/L Standard (raw)

| Content | D(+)-Glucose 0.34%(W/V) | | | |
|-------------|--|--|--|--|
| | L-Glutamic acid 0.34%(W/V) | | | |
| Preparation | Weigh 3.410g of D(+)-Glucose and 3.410g of L-Glutamic acid. Add 1L of DI | | | |
| | water and solve it. | | | |
| Waste | Dilute big amount of water and waste. | | | |

0.5M Phosphate acid buffer (raw)

| Content | Potassium Dihydrogen-Phosphate 6.8%(W/V) | | | |
|-------------|---|--|--|--|
| | Disodium Hydrogen-Phosphate 7.1%(W/V) | | | |
| Preparation | Weigh 340g of Potassium Dihydrogen-Phosphate and 355g of Disodium | | | |
| | Hydrogen-Phosphate. Add 5L of DI water and solve it. | | | |
| Waste | Dilute big amount of water and waste. | | | |

(Appendix 3) Consumption of reagents

Default setting

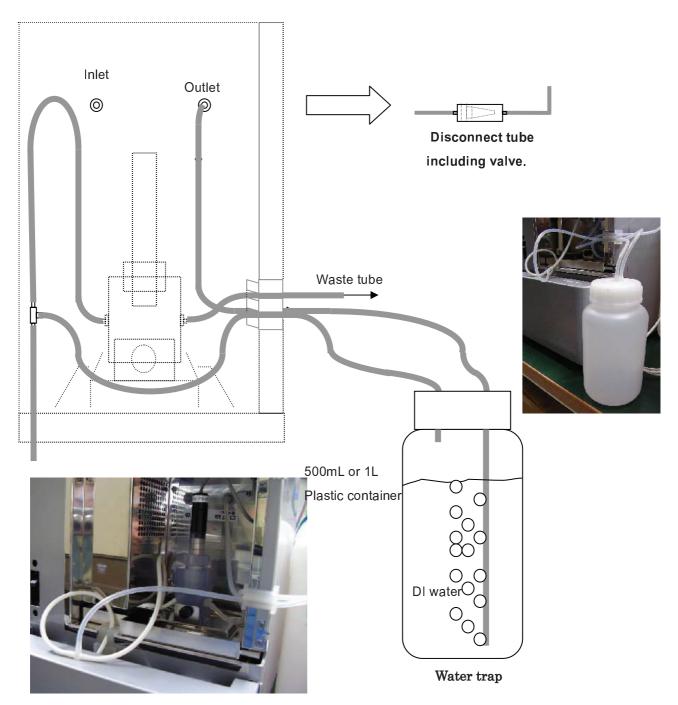
P1 rotation number: 2 (1.0mL/min.), P2 rotation number: 2 (1.0mL/min.)

| | Time of 1measurement | 24hour | 7day | 10day |
|----------|----------------------|--------|-------|-------|
| Rinse | 45min | 1.08L | 7.6L | 11L |
| Buffer | 58.33min | 1.40L | 9.8L | 14L |
| Standard | 5min | 120mL | 840mL | 1.2L |
| Sample | 8.33min(5min+200sec) | 200mL | 1.4L | 2.0L |

Notice ...Prepare enough volume of each reagents

(Appendix 4) Connecting a water trap

If some material in ambient air affect to measurement performance, it may need to connect "water trap bottle".



- Put right side of the analyzer.
- · Disconnect air filter
- Notice the direction of tube when connecting tube to water trap bottle.

| | Refer O&M manual | Neiel Oan Hallua | p.17∼18 | | | p.20 | | | p.30∼33 | p.19~29 | p.34∼38 |
|-----------------------------|-----------------------|------------------|--|--|---|--|------------------|--------------|--|--|---|
| | y. | yearly | | | | | | | | | • |
| | | 2 month | | | | | | | <u>•</u> | <u> </u> | 0 |
| | frequen | monthly | • | • | | | | • | . | <u>•</u> | |
| | Maintenance frequency | weekly | * | • | | • | 0 | | | | |
| | Mai | 2 week | | | • | • | | | | | |
| | | At use | 0 | 0 | 0 | | 0 | | (0) | 0 | |
| BOD Analyzer QuickBOD α1000 | tricodycod | Clecybolin | ① Prepare enough volume.② Wash container using DI water once per month. | ① Prepare enough volume.② Wash container using DI water once per month. | Prepare enough volume. Prepare when you use. Not prepare in advance. Use clean container. Wash container using detergent and brush; rinse it using DI water before preparation. Use clean glass tools. Dilute at exactly. | Prepare correct solution. Prepare enough volume. | Waste correctly. | No clogging. | Check the tube, joint and flow through cell. Change dirty tube and joint. Clean flow through cell. | Check the oxygen consumption ratio >10%. Measurement result should be less than 2mg/L when measuring same grade water as rinse solution. | Change membrane and electrolyte periodically. |
| | nce | Procedure | Check Refill or change | Check Refill or change | Check change | Change | Check / waste | Change | Check / change / | Check | Maintenance |
| BOD Analyzer | Maintenance | ltem | Rinse solution | Buffer solution | Standard Standard solution (for calibration) | Sample water | Waste bottle | Air filter | Tube / flow through cell | Microbe membrane | DO electrode |

□:waste

o:check

■:clean

♦:refill

:change/preparation