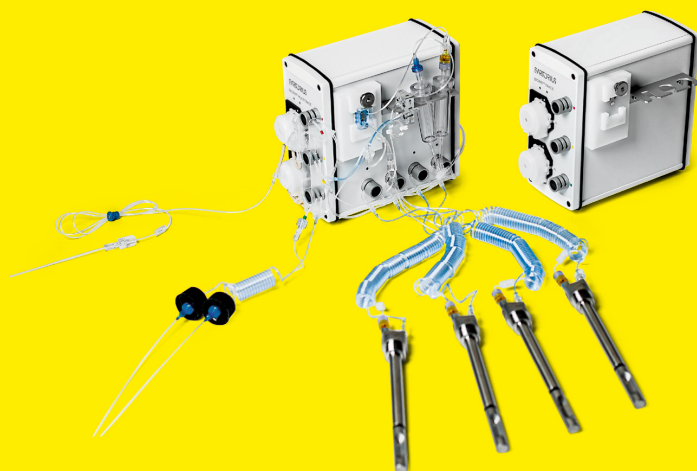


Operating Instructions

BioPAT® Trace | Multi Trace

2- | 2 × 4-Channel Online Analysis System



85032-541-53



SARTORIUS

Contents

1	About this Document	8
1.1	Introduction	8
1.2	Validity	8
1.3	Target Groups	9
1.4	Symbols Used	10
1.4.1	Warnings in Operation Descriptions	10
1.4.2	Other Symbols Used	10
2	Safety Instructions	11
2.1	Intended Use	11
2.2	Personnel Qualification	11
2.3	General Safety Instructions	12
2.3.1	Electrical Power	12
2.3.2	Energy Release	12
2.3.3	Accessories, Consumables, and Spare Parts	13
2.4	Conduct in an Emergency	13
3	BioPAT® Trace Multi Trace Product Description	14
3.1	The Basics of Electrochemical Measuring Technology	15
3.2	Biosensor Principle	16
3.3	Fluidic System and Sampling	17
3.4	Data Transmission and Data Saving	20
3.5	Equipment Supplied with the BioPAT® Trace Multi Trace	20
3.6	Consumables	21
3.6.1	BioPAT® Trace BioPAT® Multi Trace Tubing Set	21
3.6.2	Buffer Solution	21
3.6.3	Cleaning Solution	21
3.6.4	Calibration Standard Solutions (contents: 0.5 liters)	22
3.6.5	Membranes	23
3.7	Sampling Probes	23
3.7.1	Dialysis Probe	23

3.8	Accessories for Sampling Probes	24
3.8.1	Accessories for Dialysis Probe	24
3.8.2	Accessories for Dialysis Probe (1st generation, no integrated Luer-Lock connection)	25
3.9	Accessories for the BioPAT® Trace Multi Trace	25
4	Design and Function	26
4.1	Setting Up the Device	26
4.2	BioPAT® Trace Multi Trace Measurement Methods	29
4.2.1	Dialysis Measuring Method Using the Dialysis Probe	30
4.3	Conductivity Cell Function	33
4.4	Particle Filter Function	34
4.5	Diffusion Module Function	35
4.6	Dialysis Probe Function	36
4.7	Enzyme Reactor Function	37
4.8	Temperature Correction	37
5	Transport	38
6	Initial Installation	39
6.1	Setup	39
6.1.1	Device Setup	39
6.1.2	Tubing Set Connectors on the Device	41
6.1.3	Attaching the Tubing Set for the “Dialysis” Operating Mode	46
6.1.4	Installing the Tubing Set for the BioPAT® Multi Trace	54
6.1.5	Connection and Removal of the Enzyme Reactor	54
6.2	Connecting the Dialysis Probe	56
6.2.1	Mounting the Membrane in the Dialysis Probe	57
6.2.2	Mounting the Membrane for Measuring Methanol and Ethanol in the Dialysis Probe	58
6.2.3	Installing the Dialysis Probe into the Bioreactor	59
6.2.4	Sterilization of the Dialysis Probe Prior to Connection	59
6.2.5	Connecting the Dialysis Probe to the Tubing Set	61
6.3	Cleaning the Dialysis Probe and Replacing the Membrane	62
6.4	Loading the Software and Network Connection	63
6.4.1	Troubleshooting	66

6.5	Installing the Software	66
6.6	Changing the Start Settings	67
6.7	Active Access Restriction Log in.....	68
6.8	Assignment of Analog Outputs.....	69
6.9	Checklist for Starting-up Operation	70
7	Startup.....	73
7.1	Startup.....	73
7.2	Decommissioning.....	74
8	Operation.....	75
8.1	Basic Commands	78
8.1.1	Start	78
8.1.2	Standby	78
8.1.3	Calibrate	78
8.1.4	Single Sample	79
8.2	Reference (for Dialysis only)	79
8.2.1	Fast Reference (Only for Dialysis)	82
8.3	Menu Bar	83
8.3.1	Menu Item "Device"	84
8.3.2	Menu Item "Display"	87
8.3.3	Print Diagram	87
8.3.4	Calibration Values.....	88
8.3.5	Calibration History	88
8.3.6	Status.....	88
8.3.7	Settings	89
8.3.8	Menu "Schedule".....	96
8.3.9	Menu "Controller".....	100
8.3.10	Menu Item "Log in".....	105
8.3.11	Menu Item „Information“.....	105
8.3.12	Data Transfer via TCP/IP.....	108
8.4	LED Status Messages.....	109

8.5	Tips	110
8.5.1	Choosing Suitable Standard Solutions	110
8.5.2	Dialysis Mode: Referencing at the Beginning of a Fermentation	111
8.5.3	Dialysis Mode: Referencing at Low Lactate Concentrations.....	111
8.5.4	Avoiding Contaminations in the Tubing Set	112
8.5.5	Import Data to Excel	113
8.5.6	Data Transfer to Excel Using an Excel Macro	115
9	Maintenance and Servicing	116
9.1	Cleaning	116
9.2	Decontaminating the Tubing Set	116
9.2.1	Contamination by Biofilm	116
9.2.2	Causes of Contamination	117
9.2.3	Decontaminating Connection Lines	117
9.2.4	Decontaminating the Transport Buffer	117
9.2.5	Decontaminating the Tubing Set	117
9.2.6	Disposal Line Disposal Tank	118
9.2.7	Probe Sterilization.....	118
9.3	Maintenance and Servicing Activities.....	118
9.3.1	Device.....	118
9.3.2	Tubing Set.....	118
10	Malfunctions.....	119
10.1	Error messages	119
10.1.1	Calibration Error.....	120
10.1.2	Calibration Implausible.....	120
10.1.3	Sensor Error.....	121
10.1.4	Error in Control	121
10.2	Error and Possible Causes	122
10.3	Error Message: “Calibration Failed”	124
11	Transport.....	126

12 Storage and Shipping	127
12.1 Storage	127
12.2 Returning the Device and Parts	127
13 Disposal	128
13.1 Information on Decontamination	128
13.2 Decommissioning the Device	128
13.3 Disposing of the Device and Parts	129
13.3.1 Information on Disposal	129
13.3.2 Disposal	129
14 Technical Data	130
14.1 Measuring Data	130
14.2 Dimensions and Weights	130
14.3 Installation Conditions	131
14.3.1 Ambient Conditions	131
14.3.2 Footprint	131
14.3.3 Storage Conditions	131
14.4 Electrical Data	132
14.4.1 Power Supply	132
14.4.2 Interfaces	132
15 Conformity Documents	133
16 Licensing or Trademark Information	136

1 About this Document

1.1 Introduction

These instructions provide all the information necessary for operation of the BioPAT® Trace | Multi Trace (hereinafter referred to as the device).

The instructions must be read, understood and used by all personnel using the device.

- ▶ Before working with the device: Read the instructions carefully and completely.
- ▶ Keep them in a safe and easily accessible place near the device's site of installation. These instructions are part of the device.
- ▶ If the instructions are lost: Request a replacement or download the latest instructions from our website.

Masculine or feminine forms are used to facilitate legibility in these instructions and always simultaneously denote the other gender as well.

1.2 Validity

These instructions apply to BioPAT® Trace | Multi Trace in the following versions:

Device	Type
BioPAT® Trace	BPT0001
BioPAT® Multi Trace	BPT00M

In general, these instructions apply to both devices (BioPAT® Trace and BioPAT® Multi Trace). The procedural steps describe the standard tasks for the BioPAT® Trace. Differences or deviating procedural steps for the BioPAT® Multi Trace will be marked as such.

1.3 Target Groups

These instructions are designed for the following target groups. The target groups must possess the knowledge listed.

Target group	Knowledge responsibilities
User	<p>The user is familiar with the operation of the device and the associated work processes. He knows the dangers that can occur when working with the device and can avoid these dangers.</p> <p>The user has been trained in the operation of the device.</p> <p>The training takes place within the scope of startup and is carried out by the operating engineer laboratory manager or the operator of the device.</p>
Operator	<p>The operator of the device is responsible for compliance with safety requirements and workplace safety regulations.</p> <p>The operator must ensure that all persons who work with the device have access to the relevant information and have been instructed in work with the device.</p>

1.4 Symbols Used

1.4.1 Warnings in Operation Descriptions

WARNING

Denotes a danger with risk that death or severe injury may result if it is **not** avoided.

CAUTION

This symbol denotes a possible danger with risk that moderate or minor injury may result if it is **not** avoided.

NOTICE

This indicates a hazard with a low risk which could result in property damage if **not** avoided.

1.4.2 Other Symbols Used

- ▶ Required action: Describes activities that must be carried out. The actions in the sequence must be carried out in succession.
- ▷ Result: Describes the result of the activities carried out.

2 Safety Instructions

2.1 Intended Use

The device is used for online monitoring of the analytes glucose, lactate, methanol and ethanol in laboratory or industrial cultivations of micro-organisms and cell lines.

The device may only be used indoors.

These instructions are part of the device. The device is intended exclusively for use in accordance with these instructions.

Foreseeable Misuse

Any use beyond the intended use of the device can cause severe unforeseeable hazards and is the sole responsibility of the operator.

The following uses, for example, are deemed to be improper:

- Operation outside the permissible ambient conditions, e.g. extreme temperatures, chemical vapors, moisture, shock, vibration or strong electromagnetic fields
- Carrying out unauthorized modifications or other technical changes on the device
- Connecting unsuitable devices
- Installation of unauthorized items on the device

2.2 Personnel Qualification

All persons working on the device must possess the necessary knowledge (see Chapter “1.3 Target Groups”, page 9).

The activities described in these instructions are aimed at the target group of operators. If individual actions must be carried out by other target groups, this will be indicated.

2.3 General Safety Instructions

Failure to follow the safety instructions in this manual can have serious consequences, e.g. exposure of individuals to electrical, mechanical or chemical hazards.

- ▶ Before starting up the device: Carefully read the chapter entitled “Safety Instructions”.
- ▶ Only operate the device when it is safe and in perfect working order.
- ▶ Observe the maintenance intervals (Chapter “9.1 Cleaning”, page 116).
- ▶ Have any damage repaired immediately by the Sartorius Service.

2.3.1 Electrical Power

Electrical switching elements are installed in the device. Damage to the insulation or individual components can be life-threatening. Contact with parts under voltage represents a direct danger of death.

Work on and modifications to the electrical equipment of the device may only be carried out by Sartorius Service personnel. The device may only be opened by Sartorius Service personnel.

- ▶ Check the device regularly for defects, such as loose connections or damage to the insulation. The device’s housing, power cord and all connections must be undamaged.
- ▶ Note the device’s protection class (IP rating). The protection class specifies the suitability of equipment for various ambient conditions, e.g. moisture or foreign bodies (see Chapter “14.2 Dimensions and Weights”, page 130).
- ▶ Make sure that the power connection is equipped with a ground lead.
- ▶ Do not use inadequately sized power cords.
- ▶ Keep live parts away from moisture. Moisture can cause short circuits.
- ▶ If the electrical equipment is defective, immediately turn off the power supply and contact the Sartorius Service.

2.3.2 Energy Release

Energy supply lines may be incorrectly dimensioned and not protected against impermissible fluctuations and faults. This can lead to energy being released, e.g. electric surges or gases.

The power supplies used to operate the device must be designed in accordance with the technical data (see Chapter 14.2, page 130). Energy connections must be fitted with fully functional safety equipment:

- Earth leakage circuit breaker for mains connections (ground fault circuit interrupter)
- Fittings for shutting off water, compressed air or gas supplies

2.3.3 Accessories, Consumables, and Spare Parts

The use of unsuitable accessories, consumables and spare parts can be hazardous:

- Damage to the device
 - Malfunction of the device
 - Total failure of the device
- ▶ Only use accessories, consumables, and spare parts from Sartorius. Sartorius can provide information on operational quality upon request.
 - ▶ Only use accessories, consumables, and spare parts that are in technically perfect condition.

The use of accessories, consumables, and spare parts **not** approved by Sartorius is the sole responsibility of the operator.

2.4 Conduct in an Emergency

Malfunction may result in injury or property damage.

If there is immediate danger of personal injury or equipment damage, take the device out of operation immediately:

- ▶ Unplug the power cord from the device (1).
- ▶ Have any malfunctions repaired immediately by the Sartorius Service.

3 BioPAT® Trace | Multi Trace Product Description

The BioPAT® Trace | Multi Trace analysis system is used for simultaneous online monitoring of the analytes glucose and lactate in laboratory or industrial cultivations of microorganisms and cell lines.

An additional application for the measurement of ethanol or methanol is available.

The BioPAT® Trace | Multi Trace provides continual analysis independent of the type of cultivation (batch, fed-batch, continuous cultivation).

The measurement range of the BioPAT® Trace | Multi Trace extends from 0.1 to 40 g/L for glucose, from 0.05 to 10 g/L for lactate, from 0.5 to 20 g/L for methanol and from 1 to 40 g/L for ethanol.

Depending on the conditions, the measurement frequency is 30 analyses per hour maximum. Due to the device's structured design and easy-to-operate concept, the user can perform routine analysis functions after just a brief introduction.

The ambient temperature of the BioPAT® Trace | Multi Trace can lie between 15 and 35 °C due to internal temperature correction. The ambient humidity should not exceed 90%.

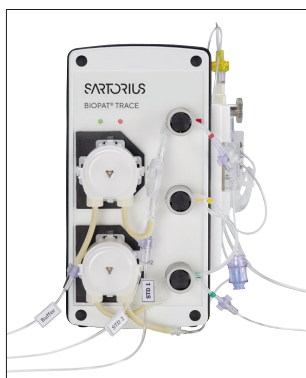
Analysis in the BioPAT® Trace | Multi Trace is a combination of enzymatic conversion and electrochemical detection. A 2-channel enzyme electrode coated with oxidase or a combination of an enzyme reactor with a platinum electrode (methanol | ethanol, (Glucose | lactate) application) serves as the biosensor. Depending on the application, the service life of the sensor system ensures 30 days or 5,000 measurements. The deviation from the average measurement is less than 3% for a measurement of 5 g/L of glucose and 2.5 g/L of lactate. For more information please request the BioPAT® Trace: Performance and Accuracy application note.

3.1 The Basics of Electrochemical Measuring Technology

An electrochemical detector measures the current produced at a specific potential applied to the electrodes by the analytes in a flow conductivity cell. In the presence of an electrolyte solution, the electrochemical conversion of the substance at the working electrode leads to the release or attraction of electrons (oxidation or reduction). The size of the measurable threshold current depends on the parameters of Faraday's law of electrolysis as well as on Fick's diffusion law and is proportional to the concentration.

The selectivity of the sensitive detector is based on the principle that not all substances are electrochemically active and thus many extraneous substances in a sample solution will not be detected. Selectivity is dependent on the working electrode's material and the applied potential.

By combining this system with enzymes, the selectivity of an electrochemical sensor can be raised even higher – and the electrode is turned into a biosensor.



3.2 Biosensor Principle

The analytes glucose and lactate are detected by means of enzymatic reactions. Figure 1 shows the enzymatic conversion of glucose to gluconolactone. In the presence of water, gluconolactone is immediately hydrolyzed to gluconic acid. The hydrogen peroxide (H_2O_2) produced in the first step is detected amperometrically through anodic oxidation that releases the two electrons. Figure 2 shows the enzymatic conversion of lactate to pyruvate. During this reaction as well, H_2O_2 is formed and then detected amperometrically.

Enzyme reactions take place according to the “key-lock principle”. They are specific and thus highly selective. Therefore, the reaction at the enzyme system is a further selection. The combination of the two selective principles makes the BioPAT® Trace | Multi Trace largely insensitive to matrix effects and extraneous materials.

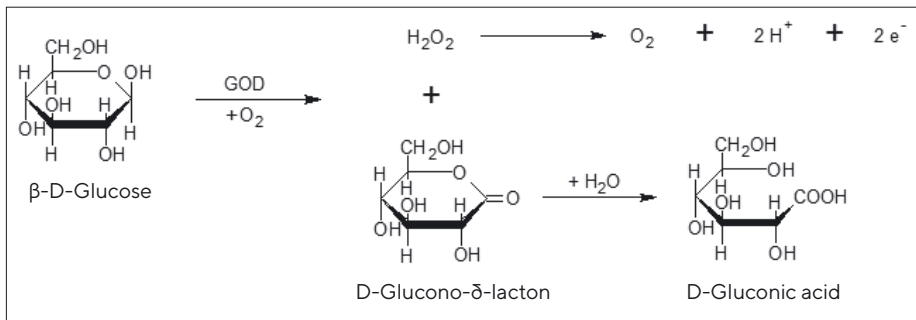


Fig. 1: Enzymatic conversion of glucose with parallel anodic oxidation of H_2O_2

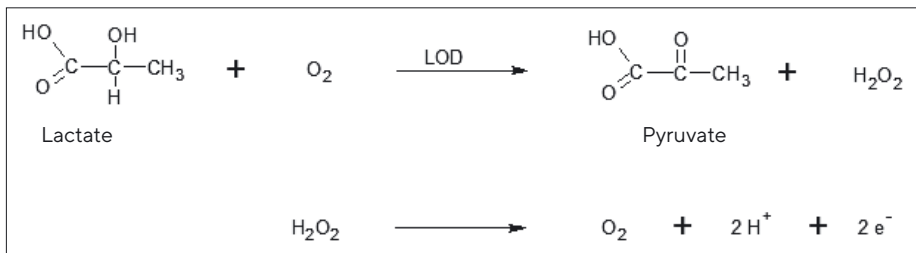


Fig. 2: Enzymatic conversion of lactate with parallel anodic oxidation of H_2O_2

The BioPAT® Trace | Multi Trace biosensors consist of platinum thick-film electrodes that are durably coated with the corresponding enzymes (glucose oxidase (GOD) or lactate oxidase (LOD)). Then, enzyme and electrode are in close proximity to each other, so that the reactions described above can be recorded directly.

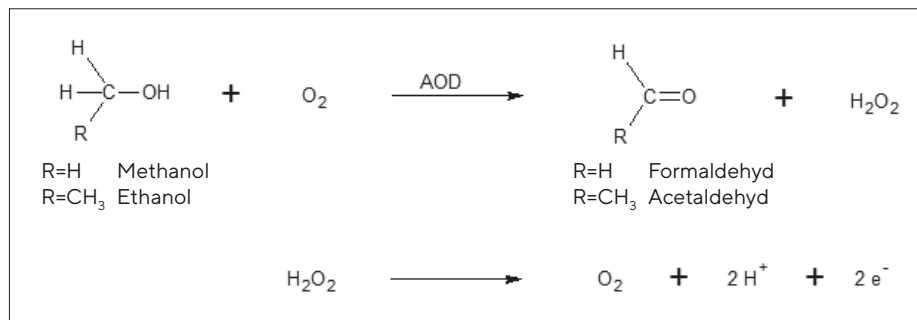


Fig. 3: Enzymatic conversion of methanol | ethanol with parallel anodic oxidation of H₂O₂

Figure 3 shows the enzymatic conversion of methanol or ethanol to formaldehyde or acetaldehyde. The enzyme alcohol oxidase (AOD), which occurs in an enzyme reactor in immobilized form, is used as catalyst. The hydrogen peroxide (H₂O₂) formed in the reaction is detected amperometrically. With this method, the alcohol is converted in the enzyme reactor and the detection of the formed hydrogen peroxide takes place on a platinum electrode.

3.3 Fluidic System and Sampling

To perform the respective analysis on a sample, representative material must be transported to the biosensor. A fluidic system (tubing set) consisting of tubing lines, pumps and valves is used for that purpose. Additionally, the sample is also conditioned in the fluidic system, i.e. potentially interfering components are removed, and diluted to the extent that it can be assayed reliably by highly sensitive biosensors. This is done by separation through a dialysis membrane, which is integrated in every tubing set as a diffusion module.

Conditioning of the sample is based on the method of flow diffusion analysis (Figure 4). This method works with a time-dependent sample transfer. For that purpose, the measurement line and the sample and/or the sample infeed are separated from each other by a membrane. The measuring distance is called the acceptor (receiving) and the sample side is called the donor (giving).

During an analysis, the measurement line is flushed with a transport buffer, which is an acceptor stream that can be stopped from behind the membrane for a defined period of time. This procedure is carried out by triggering a valve, which in turn opens up a bypass route.

Meanwhile, the analytes in the sample diffuse through the membrane and, at the same time, are enriched in the holding chamber. In this way, a continually increasing concentration of dialyzable components develops in the fluid segment of the transport buffer.

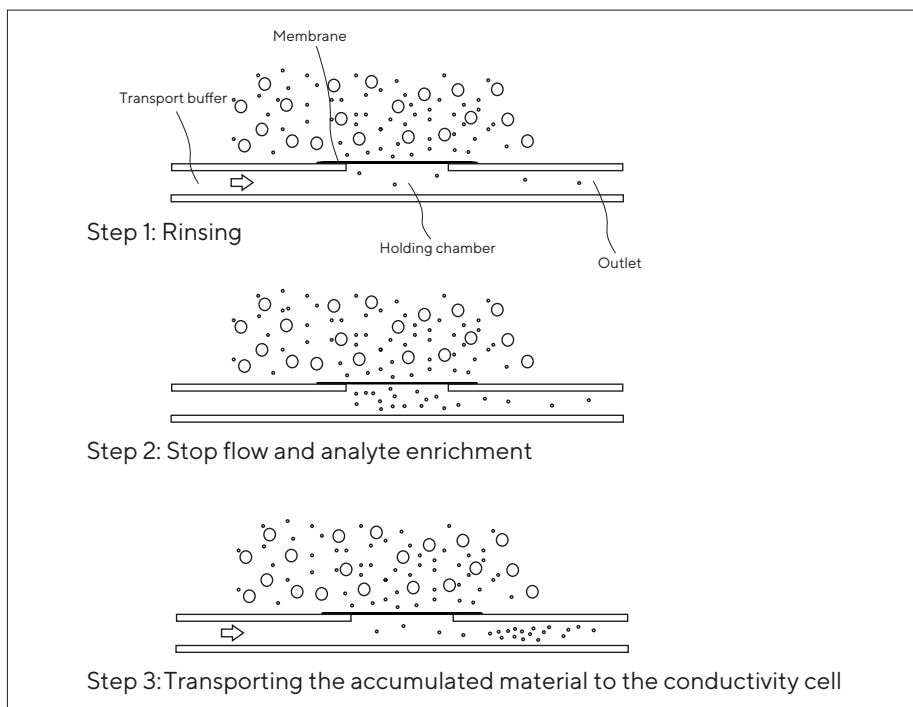


Fig. 4: Principle of the flow diffusion technique

When this segment is released by re-switching the valve, the flow goes to the conductivity cell, where it generates a signal. The intensity of the signal is a measure of the concentration of the sample. One big advantage of this enrichment method is the very broad measuring range that is achieved by varying the diffusion time; it also protects the sensor against inferring particles in the sample.

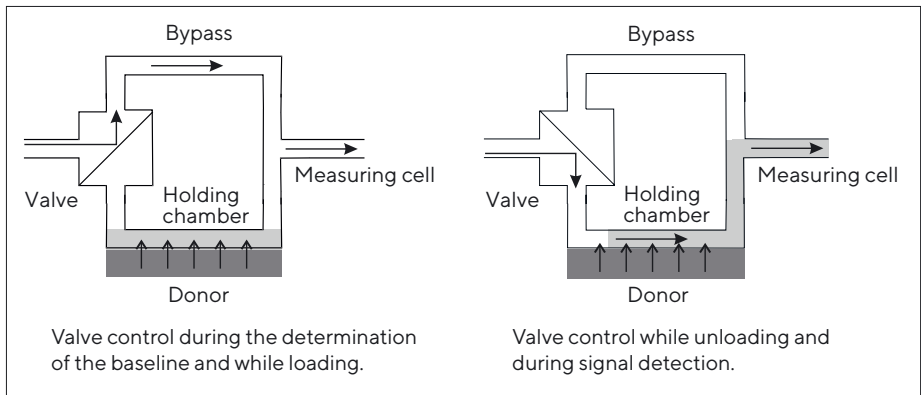


Fig. 5: Valve control

PID Control

Feeding in a cultivation can be reliably controlled with the integrated PID controller. The PID controller consists of three interacting single controller. These are the proportional controller (P), the integral controller (I), and the derivative controller (D). The control action of the controller is described by a differential equation:

$$u(t) = K_P \times e(t) + \frac{K_P}{T_N} \times \int_0^t e(\tau) d\tau + K_P \times T_V \frac{d}{dt} e(t)$$

The persistent control deviation of the proportional gain of the controller can be balanced with the integral gain. With the derivative gain of the controller it is possible to react to fast changes of concentrations inside the reactor. The PID controller is better suited for feeding processes than a simple two-level controller.

Adjusting the PID Controller

The principle of the PID controller is known since 1922 (Minorsky[1]), but useful rules for adjustment were not available before 1942 (Ziegler and Nicols[2]). Today most of the controller will be adjusted by empirical design. This design can also be used in the field of biotechnology.

Literature

[1] Minorsky, Nicolas: Directional stability of automatically steered bodies; J. Amer. Soc of Naval Engineers 34 (1922), pp. 280–309.

[2] Ziegler, J.G. and Nicols, N.B.: Optimum settings for automatic controllers; Trans. ASME, 64 (1942), pp. 759-768

3.4 Data Transmission and Data Saving

The BioPAT® Trace | Multi Trace has a number of outputs, making integration into data recording systems very flexible. Along with a standard analog output for signal ranges from 0 to 20 mA, 0 to 10 V or 4 to 20 mA, the BioPAT® Trace | Multi Trace also features an Ethernet connection that connects the device to a PC via a network. Using the software supplied, the measured results can be visualized graphically and saved (see Chapter "8 Operation", page 75).

3.5 Equipment Supplied with the BioPAT® Trace | Multi Trace

Description	Order no.
Device BioPAT® Trace with power adapter, power plugs (Europe, USA, UK), cross-over Ethernet cable, operating instructions and PC software (trace_mon)	BPT0001
Device BioPAT® Trace with power adapter, power plugs (Europe, USA, UK), cross-over Ethernet cable, operating instructions and PC software (trace_mon)	BPT00M

3.6 Consumables

3.6.1 BioPAT® Trace | BioPAT® Multi Trace Tubing Set

Description	Order no.
Dialysis tubing set, glucose application lactate	BPT0089
Dialysis tubing set (ethanol methanol)	BPT0050
Enzyme reactor for ethanol methanol application	BPT0048
Dialysis tubing set, glucose application lactate for Multi Trace	BPTOM07
Dialysis tubing set (ethanol methanol) for Multi Trace	BPTOM04
Biosensor set for glucose application lactate	BPT0091

3.6.2 Buffer Solution

Description	Order no.
Salt mixture buffer, glucose lactate application Cell culture	BPT0090
Buffer solution, methanol ethanol application; Content: 2 liter, ready to use	BPT0092
Buffer solution (for microbial cultivations), Glucose application lactate; Content: 2 liter, ready to use	BPT0093

3.6.3 Cleaning Solution

Description	Order no.
Cleaning solution Methanol ethanol concentrate: 1 liter	BPT0044A
Cleaning solution Glucose lactate concentrate: 1 liter	BPT0044G

3.6.4 Calibration Standard Solutions (contents: 0.5 liters)

Description	Order no.
Calibration standard 0.1 g/L glucose, 0.05 g/L lactate	BPT0043
Calibration standard 0.5 g/L glucose, 0.25 g/L lactate	BPT0011
Calibration standard 1 g/L glucose, 0.5 g/L lactate	BPT0010
Calibration standard 2 g/L glucose, 1 g/L lactate	BPT0009
Calibration standard 4 g/L glucose, 2 g/L lactate	BPT0008
Calibration standard 10 g/L glucose, 5 g/L lactate	BPT0007
Calibration standard 20 g/L glucose, 10 g/L lactate	BPT0041
Calibration standard 0.2 g/L methanol	BPT0051
Calibration standard 1 g/L methanol	BPT0052
Calibration standard 5 g/L methanol	BPT0053
Calibration standard 20 g/L methanol	BPT0054
Calibration standard 0.5 g/L ethanol	BPT0058
Calibration standard 2 g/L ethanol	BPT0057
Calibration standard 4 g/L ethanol	BPT0056
Calibration standard 40 g/L ethanol	BPT0055

3.6.5 Membranes

Description	Order no.
Membranes (glucose lactate, higher measuring range) for the dialysis probe (contents: 5 pcs)	BPT0024
Membranes (glucose lactate, lower measuring range for the dialysis probe (contents: 5 pcs)	BPT0024LR
Membranes (glucose lactate/cellulase-stabile) for the dialysis probe (contents: 5 pcs)	BPT0040
Membranes (ethanol methanol) for the dialysis probe (contents: 5 pcs)	BPT0047
Membranes (glucose lactate), stainless-steel bypass module PEEK (contents: 5 pcs)	BPT0074
Membranes (glucose lactate low), stainless-steel bypass module PEEK, (contents: 5 pcs)	BPT0075

3.7 Sampling Probes

3.7.1 Dialysis Probe

Equipment supplied includes installation kit, two autoclaving cap sets and operating instructions

Description	Order no.
Length of dialysis probe: 132 mm	BPT0082
Length of dialysis probe: 165 mm	BPT0083
Length of dialysis probe: 212 mm	BPT0084
Length of dialysis probe: 232 mm	BPT0085
Length of dialysis probe: 300 mm	BPT0086
Length of dialysis probe: 332 mm	BPT0087
Length of dialysis probe: 432 mm	BPT0088

3.8 Accessories for Sampling Probes

3.8.1 Accessories for Dialysis Probe

Description	Order no.
Installation kit for starting up the bypass module Stainless steel	BPT0076
Dialysis probe installation kit, 2nd generation	BPT0080
Autoclaving caps	BPT0081
Adapter 25/12 mm for the installation of the dialysis probe 165 mm in the 25 mm side port (Sartorius Safety port 52 mm) incl. O-rings	BB-38242540
Adapter 19/12 mm for installation of the dialysis probe into the 12-mm cover port (M26x1); incl. O-ring	BB-8848630
Adapter 19/12 mm for installation of the dialysis probe into the 12-mm cover port (RD28× $\frac{1}{8}$ "	BB-34104704
O-ring (outer) for adapter RD28× $\frac{1}{8}$ "	BB-39120830
O-ring (inner) for adapter RD28× $\frac{1}{8}$ "	BB-39121054
Bellows with KPC port for mounting the dialysis probe on Biostat STR® (contents: 3 pcs)	BPT0079
Replacement sterilisation loop	BPT0070
Membrane lunette (ethanol methanol) for the dialysis probe	BPT0071
Replacement adapter UNF-LUER connection for the dialysis probe	BPT0072
Connection cable for Watson Marlow pump 120U	BPT0078

3.8.2 Accessories for Dialysis Probe (1st generation, no integrated Luer-Lock connection)

Description	Order no.
Dialysis probe installation kit	BPT0045
Replacement sterilisation loop (contents: 5 pcs)	BPT0071
Replacement adapter UNF-Luer connection for the dialysis probe	BPT0072

3.9 Accessories for the BioPAT® Trace | Multi Trace

Description	Order no.
Support column	BPT0012
Stand clamp	BPT0013
Replacement knurled nut for measuring cell	BPT0026
Power adapter	BPT0027
Power cord (EU, UK, USA)	BPT0028

4 Design and Function

4.1 Setting Up the Device

The BioPAT® Trace | Multi Trace is an online analysis system in a compact metal housing. Communication with the user is driven by an external computer. As shown in the following figures, the front and side panels are equipped with the elements of the pre-assembled tubing set for analysis.

The actuator and sensor components are located on the outside of the housing. A standard or sample solution is fed into the analysis system by a pump (1 b). One of the pinch valves (3) allows a selection of the solutions to be pumped. The buffer stream is driven by the upper pump (1 a). A particle filter (2) removes smaller particles and air bubbles from the buffer stream.

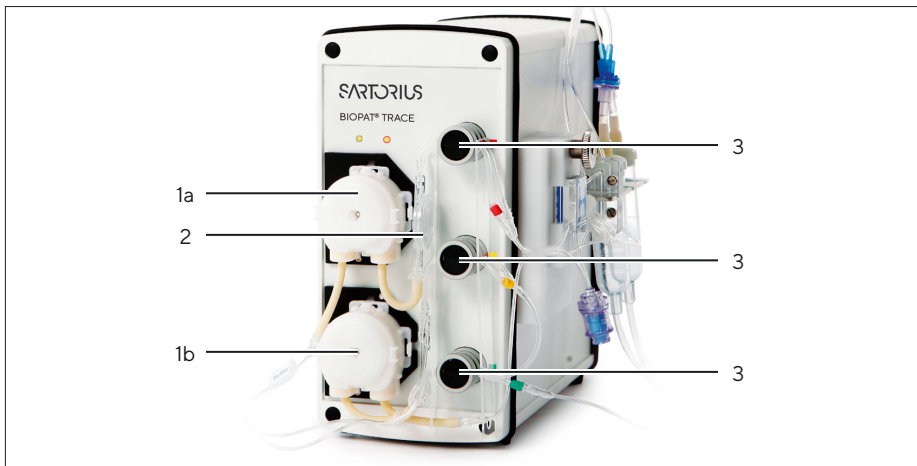


Fig. 6: Components

Pos.	Component	Pos.	Component
1a b	Pump cassettes (2 pcs.)	3	Pinch valves (3 pcs.)
2	Particle filter		

Side view of the device with the following components:

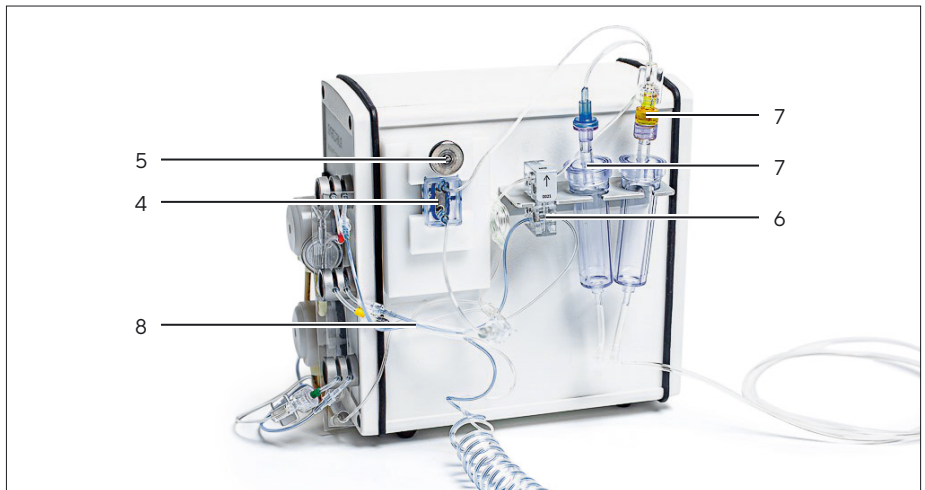


Fig. 7: Components

Pos.	Component
4	Conductivity cell
5	Knurled nut for fastening the conductivity cell
6	Diffusion module
7	Drip counter
8	Rinsing line with return valve

In the diffusion module (6), a segment of the transport buffer is enriched with the analyte from the donor stream. After time-dependent enrichment in the diffusion module (6), the acceptor stream flows into the conductivity cell (4) for assaying the analyte. The conductivity cell is fixated by a retaining plate, which can be fastened or loosened with a knurled nut (5). Furthermore, a rinsing line with return valve (8) is attached to the conductivity cell. Before the solutions are transported to waste, they each run through a drip count (7).

Rear view of the device with the following elements:

Figure 8 shows the rear view of the BioPAT® Trace | Multi Trace. The device can be switched on and off by pressing the On/off switch (9). Using the calibration data, the electrochemical signal of the biosensor is converted internally to a concentration value. The measured value is output after the analysis is completed on a PC interfaced to the Ethernet connection (10). Optionally, the signals can be output via the analog signal outputs (12). The following ranges are possible: 0..10 V, 0..20 mA, 4..20 mA. The USB interface (11) and the serial interface (RS232 port) (13) can also be used for data transfer (currently not implemented).

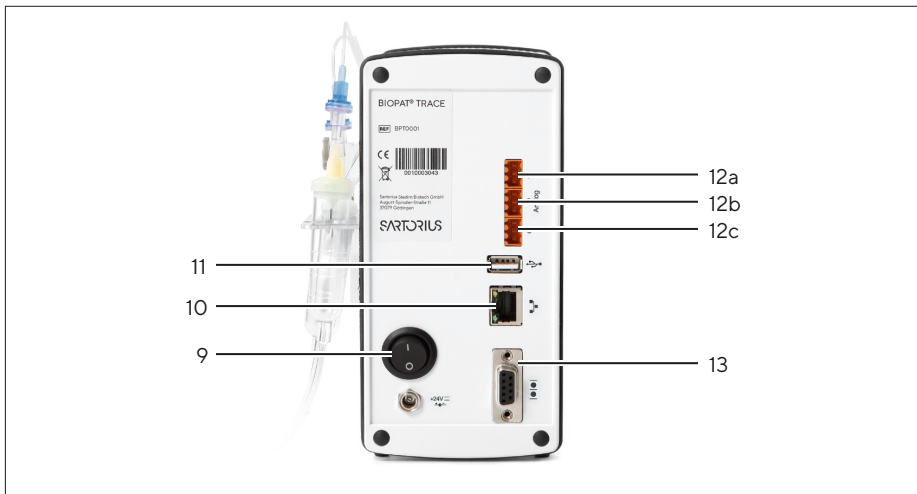


Fig. 8: Connections

Pos.	Function	Pos.	Function
9	On/off switch	12 a b c	Analog outputs (1-3)
10	Ethernet connection	13	Serial interface
11	USB interface		

The BioPAT® Trace | Multi Trace can optionally be attached to laboratory stands, suspension bars or benchtop stands. A corresponding stand clamp and a suitable benchtop stand are available as accessories.

CAUTION

Risk of damage due to incorrect connection!

Under no circumstances should a voltage source be connected to the analog data outputs at the rear of the BioPAT® Trace | Multi Trace. This also refers to a multimeter for resistance measurement. If a multimeter is used to measure the voltage or current, the contacts to the BioPAT® Trace | Multi Trace must be removed before changing the measurement range on the multimeter. Otherwise, there is a risk of the device being damaged.

4.2 BioPAT® Trace | Multi Trace Measurement Methods

BioPAT® Trace | Multi Trace enables the online sampling method of dialysis using the dialysis probe.

The simplest method is to directly measure a filtered sample of medium. However, because reactor medium is used, the range of applications is limited to processes for which there is a sufficient reactor volume or which allow continuous-feed.

Dialysis sampling is an option when processes are involved for which the reactor volume does not allow enough sample material. This method only removes low molecular substances from the reactor medium, without reducing the volume of fluid. The following figure shows a photographic representation of how the BioPAT® Trace can be connected to a bioreactor by way of a dialysis probe.

A more detailed explanation of the sampling probes can be found in Chapter “4.6 Dialysis Probe Function”, page 36.



Fig. 9: BioPAT® Trace | Multi Trace – System Setup

4.2.1 Dialysis Measuring Method Using the Dialysis Probe

The dialysis measuring method using the dialysis probe involves the transfer of analytes through a dialysis membrane in the dialysis probe which separates the reactor contents (medium) from the acceptor stream. At the same time, the dialysis membrane functions as a sterile barrier. Figure 10 shows a schematic of the flow system.

The calibration valve (green) and the three-way stopcock can be used to choose between the standards and the manual probe. The sample valve (red) and the bypass valve (yellow) can be used to select the dialysis probe and the diffusion module.

The upper part of the flow system represents the donor stream, which is responsible for the infeed of the calibration solution into the diffusion module. Depending on the task at hand, the carrier stream accumulates the analyte in the diffusion module or in the dialysis probe.

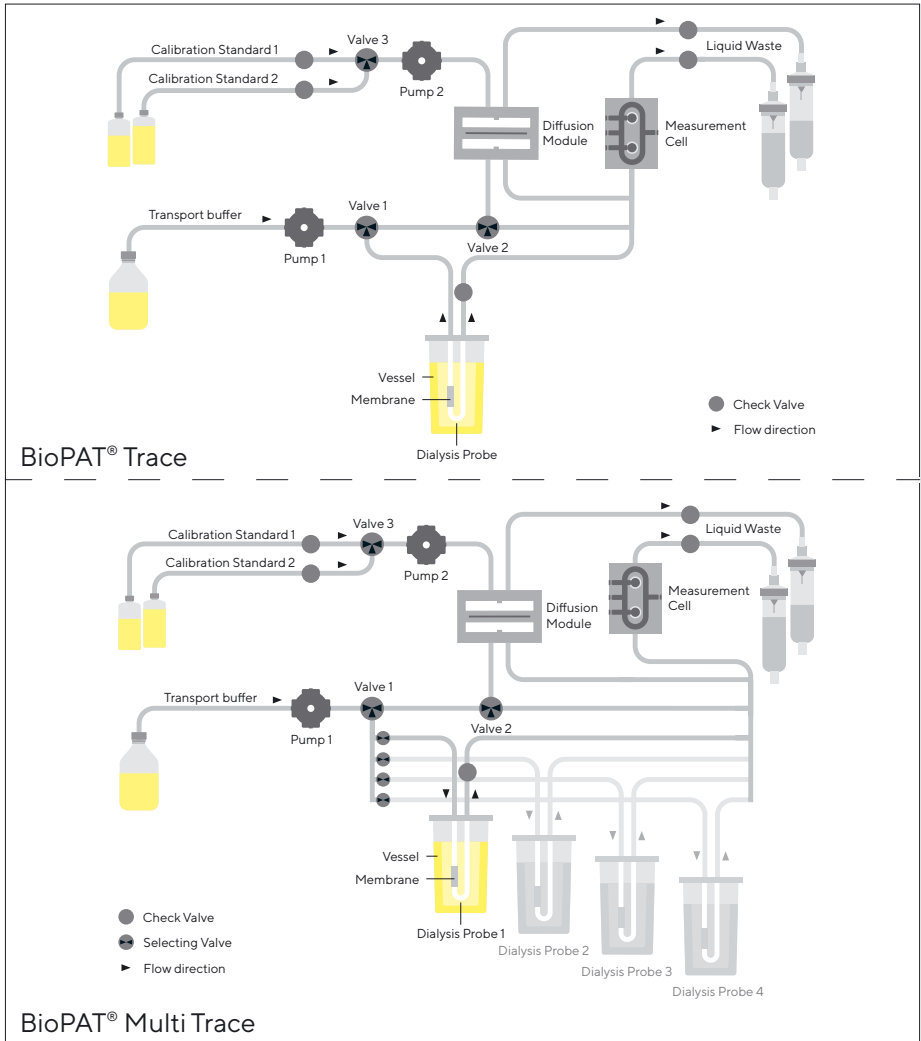


Fig.10: Flow system

For the dialysis method, the internal diffusion module is used to calibrate the BioPAT® Trace | Multi Trace. By contrast, the dialysis probe in the bioreactor is always used for measuring the sample. A reference factor between the two membranes is then determined once.

As a standard cannot be installed in the bioreactor, the BioPAT® Trace | Multi Trace must be calibrated indirectly by way of the reference factor. During this process, the initial concentration in the bioreactor is used as the starting concentration for the dialysis probe. Using the sample measurement values with an identical concentration, the BioPAT® Trace | Multi Trace calculates the factor between the diffusion module membrane and the dialysis probe membrane.

Hence, the calibration procedure for the dialysis method consists of three steps in total:

Step 1

Recording the calibration curves on the diffusion module with standards 1 and 2.

Step 2

Measuring the external sample on the BioPAT® Trace | Multi Trace with the help of the calibration curves (alternatively: by directly entering the concentration values, if known from the initial weight or by external analysis).

Step 3

Recording the calibration curves of the dialysis probe using the probe.

After measuring the standards for calculating the calibration curves, the BioPAT® Trace | Multi Trace computes a factor from the gradients of each individual curve. For all measurements carried out with the dialysis probe, this factor is applied to calculate the true concentration.

4.3 Conductivity Cell Function



The conductivity cell contains biosensors for glucose and lactate or methanol/ethanol. The conductivity cell is shown in the figure on the left. The fluid inlet flow runs perpendicular to the 2-channel sensor (“wall jet” principle). It is possible to check that the flow through the cells is free from air bubbles by using the viewing window. Any air bubbles can be removed as necessary by rinsing the measuring chamber. For that purpose, the fluid is injected through the rinsing line equipped with a return valve.

Contact springs intercept the electric signal measurable at the electrodes and transmit it to the BioPAT® Trace | Multi Trace. The conductivity cell is fastened to the device with a knurled nut.

⚠ CAUTION

Inaccurate measurement result due to air bubbles!

It must be ensured that there are no air bubbles in the measuring chamber of the flow cell, as they can distort the results.

After connecting the sampling probe and before starting each measurement series, check the conductivity cell for air bubbles through the viewing window.

As a general rule, air bubbles in the tubing set are flushed out over time. However, this process takes a certain amount of time (e.g. during the priming process).

If air bubbles are present in the conductivity cell after connecting the dialysis probe, then it would be advantageous to operate the system in measurement mode with maximum measuring frequency for a period of approx. 30 minutes.

⚠ CAUTION

Erroneous calibrations due to air bubbles!

If there are air bubbles in the measuring cell, under no circumstances should calibrations or reference measurements be carried out, as these may be erroneous and subsequently lead to incorrect results.

4.4 Particle Filter Function

The particle filter is part of the tubing set. It fulfills two tasks in the BioPAT® Trace | Multi Trace. First, sterile filtration of the buffer solution is performed through the membrane with a pore size of 0.22 µm. Before the solution leaves the module, a second membrane is used for degassing. Thus, the particle filter ensures that the acceptor stream in the BioPAT® Trace | Multi Trace stays free of particles and air bubbles.

- ▶ To ensure proper operation of the measuring instrument, the tubing set, including the particle filter, must be replaced regularly.

4.5 Diffusion Module Function

The diffusion module is part of the tubing set. The standard solutions or an external sample are pumped through the diffusion module to the analysis system. A membrane enables diffusion of the substrate from the donor solutions (both standard and sample) into the buffer solution (acceptor). The different membrane materials enable dialysis, cellulase-stable dialysis and gas diffusion for separating substrates. The following figure shows a schematic diagram of this system:

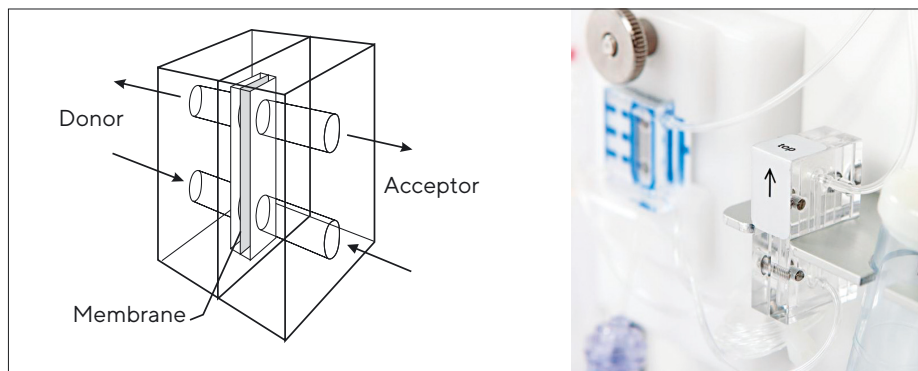
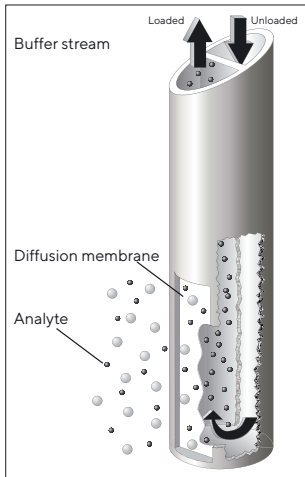


Fig.11: "Schematic diagram of the diffusion module system"

The module is made of PMMA. It has a diffusion area of 20 mm² and ensures that a sufficient exchange of substances takes place between the donor and the acceptor stream. The donor and acceptor streams flow in through the lower module inlet. The drain is located at the top.

- ▶ In order to ensure reliable operation of the measuring instrument, the diffusion module must be secured in the correct position on the retaining plate (arrow pointing up).

4.6 Dialysis Probe Function



If a dialysis probe is used to feed the sample from the bioreactor into the BioPAT® Trace | Multi Trace, then the analyte is transferred through a diffusion membrane into an internal pump-driven buffer stream and transported to the conductivity cell. The opposite figure shows the principal function of the probe.

The dialysis probe is suitable for all types of cultivation. The use of an extremely robust diffusion membrane ensures a high level of sterility. The different membrane materials enable dialysis, cellulase-stable dialysis and gas diffusion for separating substrates. The probe can be installed in the bioreactor as well as in the cover or in a side nozzle.

The application options for the dialysis probe depend on the reactor dimensions and the process conditions. In general, the dialysis probe is recommended for use on smaller bioreactors, where any loss of volume during sampling is unacceptable. The use of the dialysis probe as a sampling system ensures analysis at constant volumes.

4.7 Enzyme Reactor Function



The enzyme reactor is required for measuring methanol and ethanol. Inside the reactor is the immobilized enzyme alcohol oxidase (AOD). There, the corresponding alcohol is converted and hydrogen peroxide is formed. The hydrogen peroxide is then detected in the conductivity cell.

⚠ CAUTION

High temperatures can destroy the enzyme!

Due to the stability of the enzyme used, the ambient temperature may not exceed 25 °C. At higher temperatures, it is obligatory to use a climatic chamber.

4.8 Temperature Correction

The BioPAT® Trace | Multi Trace is equipped with a temperature sensor. This is arranged in the proximity of the conductivity cell. All measured values are corrected by using a temperature-dependent function.

5 Transport

The BioPAT® Trace | Multi Trace including accessories is delivered in protective packaging. Please save this packaging; proper shipping is only possible in the original packaging.

Upon receipt, check the delivery for completeness and any possible damage that may have occurred in transit. Any transport damage should be reported within a week of delivery. Complaints made after this date will not be accepted.

6 Initial Installation

6.1 Setup

6.1.1 Device Setup

The BioPAT® Trace | Multi Trace must be setup near the bioreactor you want to monitor (at max. distance of 2 m). The location must be free of vibrations and protected against corrosive atmospheres and chemical contamination. Additionally, the BioPAT® Trace | Multi Trace should be protected against exposure to direct sunlight.

WARNING

Risk of electric shock due to incorrect connection!

Operating the equipment with the wrong supply voltage and | or the wrong fuses can cause the danger of an electric shock.

Follow the instructions and requirements listed below concerning the power connection.

The AC adapter supplied with the device is equipped with a power plug, a mains adapter and a plug to the device. Insert the device plug into the connection socket on the rear panel of the BioPAT® Trace | Multi Trace and connect the other end to your mains outlet. The device can be switched on and off by pressing the On/off switch. When the device is switched on, a green LED lights up on the front panel.

To connect the BioPAT® Trace | Multi Trace to the local network, plug the Ethernet cable into the Ethernet connection on the back panel of the device. The operating software supplied is used to control the device via a PC. Likewise, this PC must also be connected to the local network or directly to the device using the cross-over Ethernet cable supplied.

After connecting the device to the PC, check to make sure that the system times of the two units are synchronized. Proceed as described in Chapter “8.3.7.3 Basic Settings”, page 95.

6.1.1.1 Preparing the Transport Buffer

The transport buffer is a salt mixture (for cell cultures) or a concentrate (for microbial cultures) and must be dissolved or diluted with water (deionized or distilled) according to the instructions. Proper dilution can be checked by measuring the conductivity of an aliquot. The diluted buffer must have a conductivity of 5–15 mS/cm.

To avoid contamination in the tubing set, it is recommended that the diluted transport buffer be sterile filtered and/or placed in sterile vessels such as bottles or bags. Safety measures to avoid contamination in the tubing set are summarized in Chapter “8.5.4 Avoiding Contaminations in the Tubing Set”, page 112.

CAUTION

Erroneous measurements due to incorrect or no dilution!

The transport solution is delivered as a salt mixture or concentrate. Observe the note for proper resolution/dilution on the packaging.

The consumption of transport buffer depends on the measuring interval and the number of measuring points when it is in operation. For a typical single-channel measurement in dialysis mode, the following consumption can be expected:

Dialysis mode

Interval	Buffer consumption			10 L for:	
	µl/min	ml/h	ml/d	h	d
1	835.6	50.1	1,203.2	199	8
5	835.6	50.1	1,203.2	199	8
10	614.4	36.9	884.7	271	11
20	356.3	21.4	513.1	468	19
30	270.3	16.2	389.3	617	26
60	184.3	11.1	265.4	904	38

Dialysis mode					
Interval	Buffer consumption			10 L for:	
	min	µl/min	ml/h	ml/d	h
120	141.3	8.5	203.5	1,179	49
180	127.0	7.6	182.8	1,313	55
360	112.6	6.8	162.2	1,480	62

CAUTION

Erroneous measurements due to tubing sets not being filled or only being partly filled!

Before filling the tubing set, make sure that tubing is connected to all solutions and that the vessels are sufficiently filled.

6.1.2 Tubing Set Connectors on the Device

BioPAT® Trace | Multi Trace is equipped with the corresponding tubing set (fluidics).

You will find the corresponding instructions for attaching the tubing set below.

The tubing set consists of the components necessary for analysis: Pump cassettes, valve tubes, diffusion module, drip counters and connection lines for buffers, standards, waste as well as the sampling probe (dialysis probe, dialysis module).

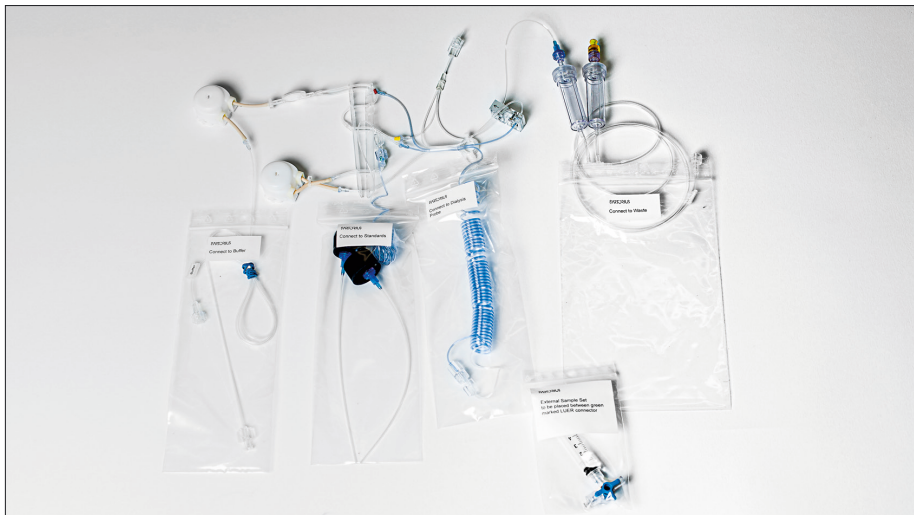


Fig. 12: BioPAT® Trace tubing set dialysis

In addition, the dialysis tubing set is provided with a single-use syringe with three-way stopcock for analyzing external samples.

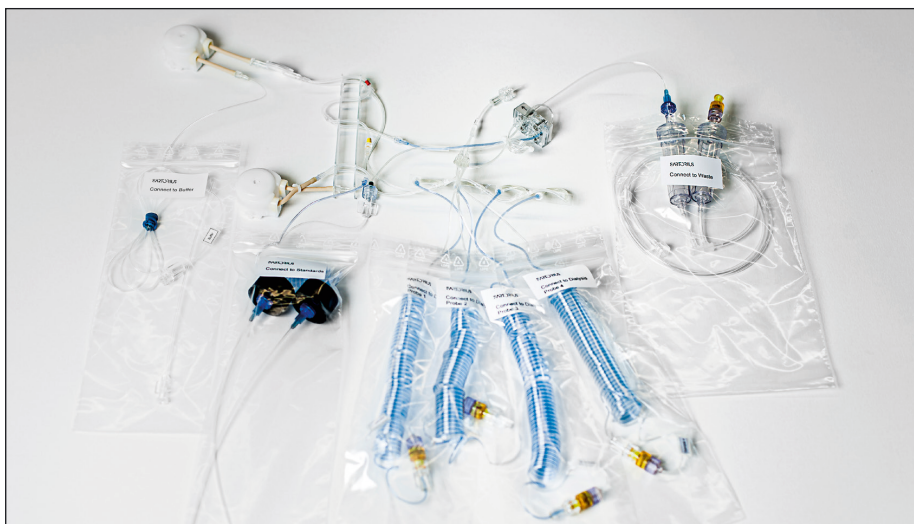


Fig. 13: BioPAT® Multi Trace tubing set dialysis

The connectivity cell with the built-in biosensor is supplied separately. It must be connected at the designated points (see arrows in Figures 12 and 13) using Luer connectors. The connection is protected from becoming mixed up by one male and one female Luer connector.

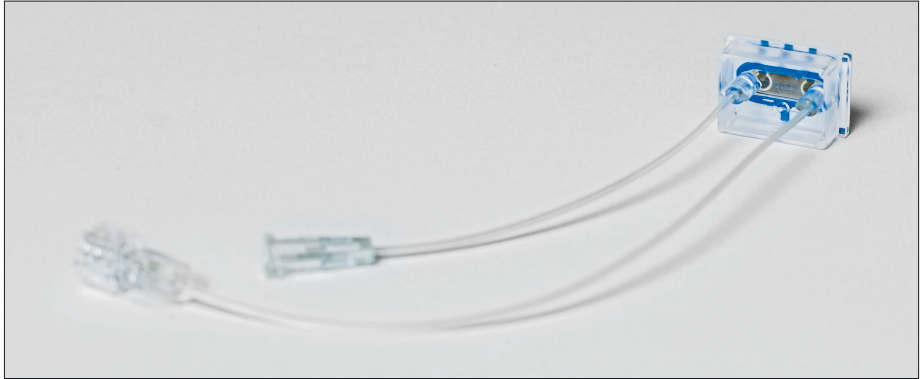


Fig. 14: Biosensor



Fig. 15: Connectivity cell connection

⚠ CAUTION**Line Blockage**

When connecting the connectivity cell, it is essential to ensure that the tubing line protruding from the male connector on the tubing set is carefully inserted into the female connector of the connectivity cell. Otherwise, the line may become blocked, leading to incorrect measurement results!

The lines for buffers, standards, sampling probe and waste are premounted and packaged in foil bags. It is recommended to only open these after the tubing set has been attached to the device in order to facilitate handling.

- ▶ Before you start attaching, remove the two empty cassettes on the pump slots. The empty cassettes can be reused as transport protection for the motor axle.

NOTICE

Only use tubing from the prefabricated original tubing set. To ensure that all solutions flow freely, never allow tubing segments to be crushed or kinked.

NOTICE

Make sure that the valve tubes are firmly seated in their pinch valves in order to ensure that the tubing can be opened and closed without problems.

The inlet and outlet lines of the tubing set are packaged in foil bags and labeled. The individual tubes are affixed with the following labels:

- BUFFER Connection to the transport buffer
 - WASTE Connection to waste
 - ← Probe port 1 dialysis
 - → Probe port 2 dialysis
 - Standard1 Standard1 Cover label for standard calibration solution 1
 - Standard2 Standard2 Cover label for standard calibration solution 2
-

CAUTION

Erroneous measurements due to tubing sets not being filled or only being partly filled!

Before filling the tubing set, make sure that tubing is connected to all solutions and that the vessels are sufficiently filled.

CAUTION

Erroneous measurements due to incorrect or no dilution!

The transport solution is delivered as a salt mixture or concentrate. Observe the note for proper resolution/dilution on the packaging.

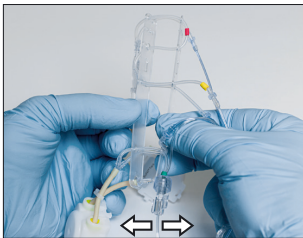
6.1.3 Attaching the Tubing Set for the “Dialysis” Operating Mode

Before attaching the filtration tubing set, the valve tubes must first be inserted into the pinch valves. To keep them from getting mixed up, each line is color-coded.

The following rules apply:

- The pair of tubes are color-coded (red, yellow and green) and are kept in place by two guidance plates
- The top-to-bottom order follows the traffic light principle: red at the top, yellow in the middle, green at the bottom
- The color-coded line of the pair of tubes is always inserted into the front valve slot
- When inserting the tubing, always guide the labeled side with your right hand

After you have inserted the tubing set, check that the result matches this picture.



As the first step, take both guidance plates in both hands and open them slightly in order to align the valve tubes in parallel.

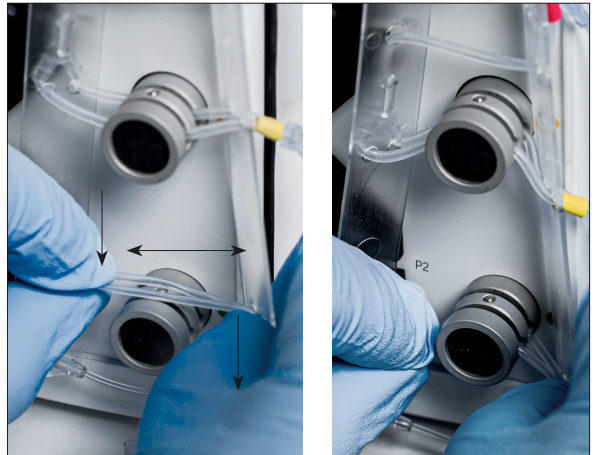
The pair of tubes marked in red should be on top and the label on the right should be visible.

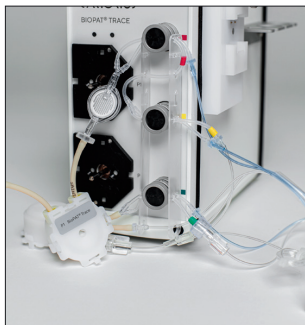


Now place the guidance plates on the valves such that the pair of tubes are positioned on their corresponding valve slots.

When inserting the tubing, make sure that the red, yellow and green color coding is always located to the right of the valve and matches that valve's color coding.

During insertion, hold both sides of the tubing taut and press the pair down until both tubes are securely seated in their slots (see figures, below). While doing this, press the black buttons on the valve alternately so that you can also insert the rear tubing deep into the valve slot. Then check that the tubes are fitted correctly in the valve in order to avoid measuring errors.





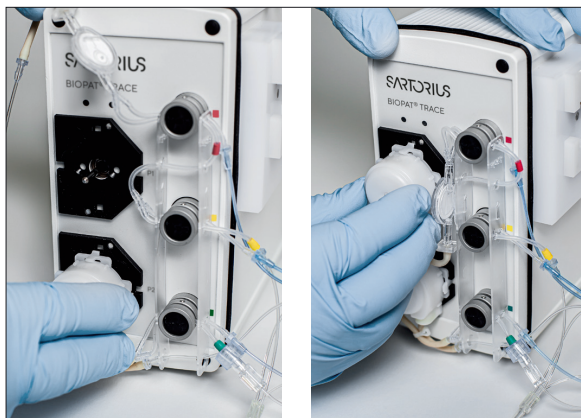
After you are done fitting all three valves, push both guidance plates towards the valve in order to secure the position of the tubes in the valve slot.

As the next step, install the two pump cassettes on the motor shaft and press on the retaining plate firmly until you hear an audible click indicating that it has locked in.

⚠ CAUTION

Before installing the pump cassettes, check if the motor shaft is clean (if necessary, wipe with a soft cloth). Dirt particles or abrasion from previous applications can adversely affect or even completely prevent the proper functioning of the pump.

The lower pump (P2) and the upper pump (P1) are attached one after the other:





Verify the proper placement of the pump cassettes.

After installing the pumps and valves on the front of the device, attach the sensor, drip chambers and diffusion module to the side panel.



The adjacent figure shows a completely assembled side panel.

First, insert the conductivity cell into the measurement amplifier holder:



- ▶ To do so, unscrew the knurled nut on the upper retaining plate to allow it to be pushed upwards.
- ▶ Next, place the lower edge of the conductivity cell into the recess in the lower border of the retaining plate.



- ▶ Leverage the conductivity cell up onto the contact plug. While doing so, pull the upper retaining plate slightly upwards.
- ▶ Press the conductivity cell onto the holder and slide the retaining plate downwards.

The conductivity cell is fastened to the device by tightening the knurled nut.

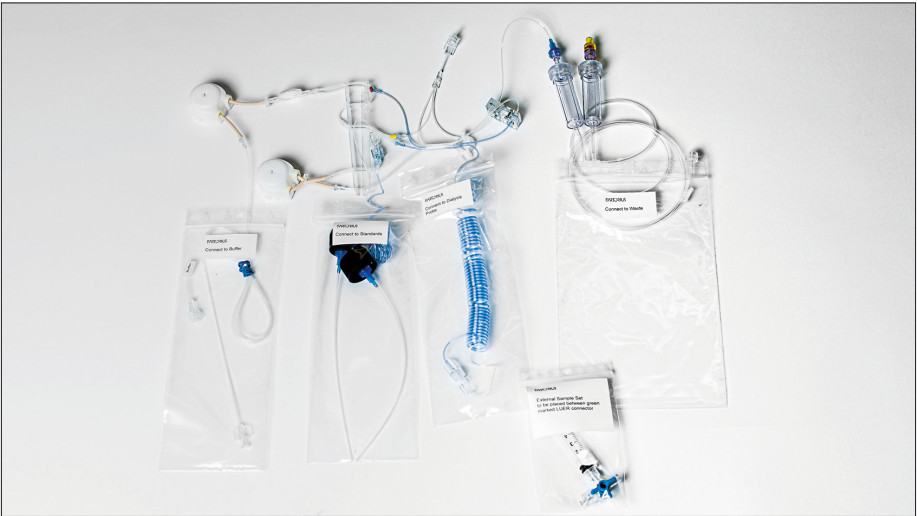


Fig. 16: BioPAT® Trace tubing set dialysis

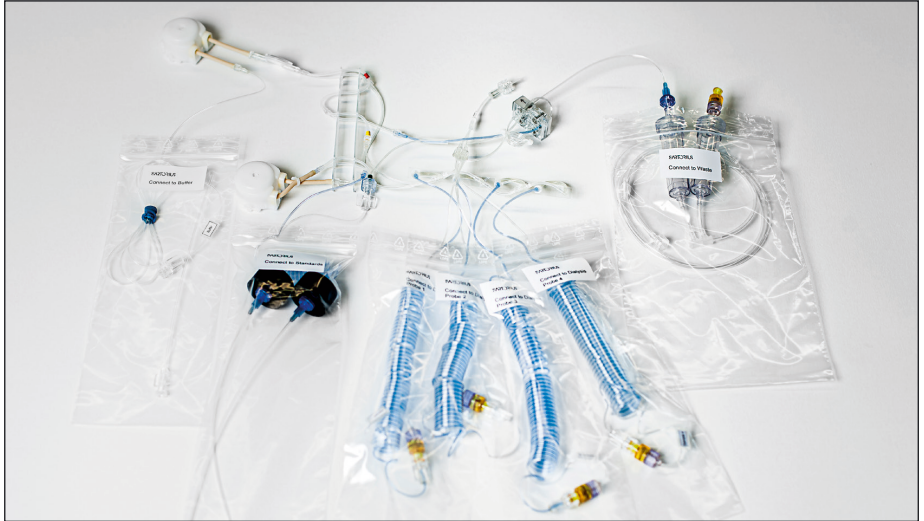


Fig. 17: BioPAT® Multi Trace tubing set dialysis

The connectivity cell with the built-in biosensor is supplied separately. It must be connected at the designated points (see arrows in Figure 1) using Luer connectors. The connection is protected from becoming mixed up by one male and one female Luer connector.

First, connect the female Luer connector to the male Luer connector on the tubing set.

⚠ CAUTION

Line Blockage!

When connecting the connectivity cell, it is essential to ensure that the tubing line protruding from the male connector on the tubing set is carefully inserted into the female connector of the connectivity cell. Otherwise, the line may become blocked, leading to incorrect measurement results!



- ▶ Afterwards, the rest of the components must be attached to the device. Using the holding slit on the side, fasten the diffusion module to the mounting rail.
- ▶ Make sure that the diffusion module is properly aligned with the arrow pointing upwards.

The conductivity cell is fastened to the device by tightening the knurled nut.



- ▶ Next, insert the two drip counters into the holders designed for this purpose.



Fig. 18: Waste line connection

Finally, the connectivity cell line (male Luer connector) is connected to the orange safety valve on the drip chamber of the waste line.

Additional Information

In addition, the dialysis tubing set is provided with a single-use syringe with three-way stopcock for analyzing external samples.

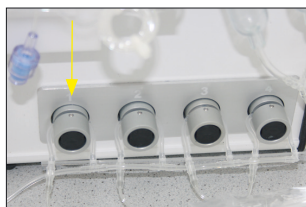
If you want to use this option, you have to place the three-way stopcock between the connection with the green label and the line to calibration standard 1.



6.1.4 Installing the Tubing Set for the BioPAT® Multi Trace



The Multi Trace tubing set is inserted in the same way as the steps described previously (for BioPAT® Trace).



Then the additional tubes are mounted.

6.1.5 Connection and Removal of the Enzyme Reactor

In the methanol/ethanol operating mode, an enzyme reactor with immobilized alcohol oxidase is required for enzymatic conversion of the analytes. Since the enzyme is relatively heat sensitive and cannot be stored dry, the reactor is delivered separately from the tubing set. The enzyme reactor must be installed into the tubing set intended for this purpose. Due to the storage conditions at 4–8°C, this should only be carried out directly prior to initial operation.

NOTICE

If air enters into the enzyme reactor, there is the danger of a decrease in the capacity of the enzyme reactor resulting in lower sensitivity.

Prior to installing the reactor, the tubing set must be inserted into the BioPAT® Trace | Multi Trace and filled with buffer. Dry setup may reduce the reactor capacity and should therefore be avoided under all circumstances. First, start the filling process to fill the tubing set (see Chapter 7, page 73).

After all lines have been filled with transport buffer, a status window opens and prompts you to install the enzyme reactor (see Figure 19).

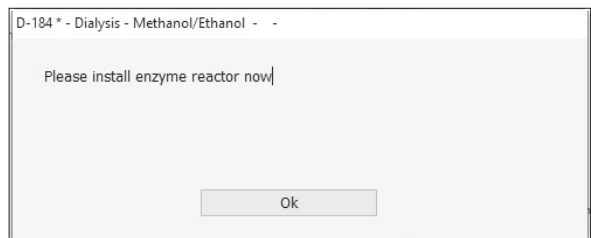


Fig.19: Enzyme reactor installation prompt

In order to install the enzyme reactor, remove it from the transport container and open the red cover caps. Afterwards, install it into the intended location in the tubing set.



After installing the enzyme reactor, follow the instructions on the monitor and the filling process will continue with the installed enzyme reactor.

NOTICE

For further usage, the enzyme reactor must be removed from the tubing set, filled with transport buffer and stored in the refrigerator at 4–8°C.

If air enters into the enzyme reactor, there is the danger of a decrease in the capacity of the enzyme reactor resulting in lower sensitivity.

The removal of the reactor is also carried out in “stopped” status (“Device – Stop”). The reactor is closed with the red cover caps, placed into the transport container and stored at a cool temperature between 4–8°C.

6.2 Connecting the Dialysis Probe

⚠ CAUTION

Erroneous measurements due to improper assembly and installation!

Ensure that the following tasks are carried out with the utmost care. Only properly assembled and installed components in perfect condition enable a trouble-free working probe.

6.2.1 Mounting the Membrane in the Dialysis Probe

Install a new membrane before every online measurement involving bioprocesses with sterile feeds as shown in Figure 14. If this rule is not observed, a lack of sterility may jeopardize the entire cultivation process.



Fig.20: Dialysis membrane assembly

Pos.	Assembly Components
1	Membrane sandwich
2	Probe lunette
3	TORX screws, M3
4	Lower part of the dialysis probe

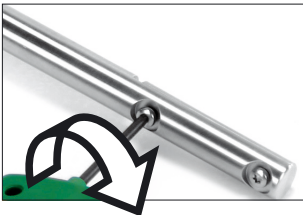
To assemble, proceed as follows:



- ▶ Insert the new membrane into the lower part of the dialysis probe. Ensure that the white side of the membrane points in the direction of the dialysis probe (in contact with the buffer) and the colored side of the membrane points in the direction of the probe lunette (in contact with the medium).



- ▶ Mount the probe lunette on the upper part of the dialysis probe.



- ▶ Attach the dialysis probe to the lower part of the dialysis probe with the supplied TORX key.

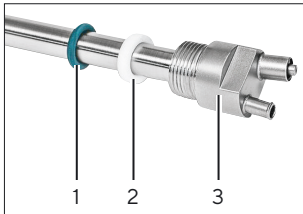
6.2.2 Mounting the Membrane for Measuring Methanol and Ethanol in the Dialysis Probe



This special probe lunette must be used to measure methanol and ethanol. This is the only way to prevent obstructions at the membrane lunette and to avoid incorrect readings or leakages in the tubing set.

- ▶ Use the special probe lunette with identifier “-0.2”.

6.2.3 Installing the Dialysis Probe into the Bioreactor



- ▶ Fit the friction washer (2) and O-ring (3) to the dialysis probe (3).

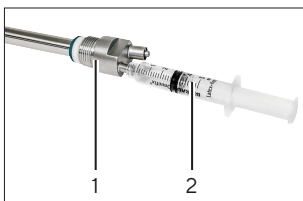
NOTICE

Note the 5-cm minimum immersion depth of the dialysis probe in the bioreactor, otherwise this could lead to incorrect readings.

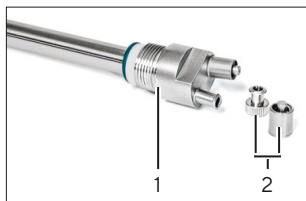
- ▶ Install the dialysis probe on the bioreactor.

6.2.4 Sterilization of the Dialysis Probe Prior to Connection

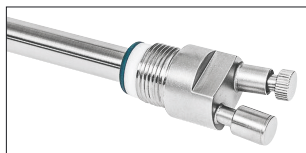
Before connecting the dialysis probe to the measuring instrument BioPAT® Trace | BioPAT® Multi Trace), the probe must be sterilized along with the bioreactor.



- ▶ Connect the dialysis probe (1) to a single-use syringe (2).
- ▶ Fill the dialysis probe with deionized water using the single-use syringe.
 - ▶ At the same time, check the dialysis probe for leaks.
- ▶ Remove the single-use syringe.



- ▶ Seal the dialysis probe (1) with the cover caps (2). This will prevent one-sided overpressure from occurring at the membrane in the dialysis probe, which could damage the membrane.



- ▶ Sterilize the dialysis probe

NOTICE

Dry sterilization is not suitable, as the membrane can become damaged.

Steam sterilization at 1 bar and 121°C. The prerequisite is that the membrane has been properly installed.

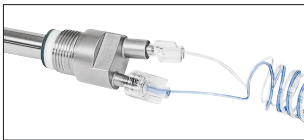
During sterilization, the dialysis probe must be filled with deionized water and sealed using cover caps, as the membrane can become damaged.

- ▶ Sterilize the dialysis probe and the bioreactor.
- ▶ Allow the dialysis probe and bioreactor to cool down sufficiently after sterilization.
- ▶ Fill the dialysis probe with deionized water using the single-use syringe.
- ▶ Check the ionized water for free flow.
- ▶ Now, the dialysis probe can be connected to the corresponding tubing set of the online analyzer.

6.2.5 Connecting the Dialysis Probe to the Tubing Set

When connecting the probe to the tubing set, ensure that as little air as possible enters the tubing set, as this is in the form of small air bubbles, e.g. could get stuck in the measuring cell and lead to fluctuating readings.

Connect the dialysis probe directly with the dialysis tubing set of the online analyzer.



- ▶ Rinse the probe with deionized water.
- ▶ First, only connect the supply line (arrow in the direction of the probe). Leave the outlet line (arrow away from the probe) free.
- ▶ Start the measurement.
- ▷ This triggers the rinsing of the probe and liquid and, if necessary, residues of air to escape from the probe outlet line.
- ▶ Connect the second line of the tubing set and continue flushing for a few minutes.

NOTICE

If there are air bubbles in the measuring cell, under no circumstances should calibrations or reference measurements be carried out, as these may be erroneous and subsequently lead to incorrect results.

As a general rule, air bubbles in the tubing set are flushed out over time. However, this process takes a certain amount of time (e.g. during the priming process). If there are air bubbles in the measuring cell after connecting the dialysis probe, then proceed as follows:

- ▶ Operate the system for a period of 30 minutes in measurement mode with maximum measuring frequency.

6.3 Cleaning the Dialysis Probe and Replacing the Membrane

After harvesting the bioreactor or any dead autoclaving phase, proceed as follows:

- ▶ Loosen the tube ends and disassemble the tubing set from the probe.
- ▶ Remove the dialysis probe from the bioreactor.
- ▶ Use a soft brush and water to clean the surface of the probe and cover caps.
- ▶ Check the gaskets of the dialysis probe adapter for cleanliness and ensure that they are seated correctly.
- ▶ Unscrew the two TORX screws from the lower part of the dialysis probe.
- ▶ Disassemble the dialysis probe lunette with the membrane from the lower part of the dialysis probe.
- ▶ Clean the dialysis probe lunette and the lunette holder with a soft brush and water.

NOTICE

If the residues are not removed, the dialysis probe may become obstructed and leakages may occur in the tubing set.

- ▶ Remove the residue from inside the dialysis probe.
 - ▶ Flush the inside of the dialysis probe with deionized water. Use the single-use syringe for this.
 - ▶ Air dry the inside of the dialysis probe. Use the single-use syringe for this.

6.4 Loading the Software and Network Connection

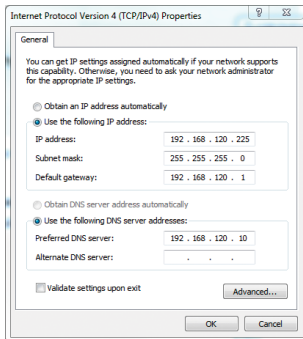
The BioPAT® Trace | Multi Trace is run internally by a computer and can be operated as a full-ranked member in intranets and LANs, or can be directly interfaced with a PC.

In order to do this, the operating software supplied must be installed on the designated PC.

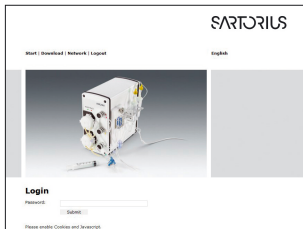
The files needed for the installation of the operating software trace_mon are stored in the device's data memory and can be accessed via an internal device website.

To install the software, proceed as follows:

- ▶ Connect the BioPAT® Trace | Multi Trace to your network and navigate to the factory-set IP address 192.168.120.231 in your web browser. Here you have the option of downloading important files and the operating instructions onto your computer and of assigning the device an IP address appropriate for your own network.
- ▶ If you do not want to make any changes (if there is a direct connection with a PC via a cross-over Ethernet cable, for example), then the device will work with the factory-set IP address. Make sure that your PC is not set to automatic address assignment when using a direct connection!



- ▶ First download the other important files for the rest of the installation onto your computer by clicking the right mouse button. They can be found in the “Download” menu item. The following files can be found here:
 - setup_tracemon_x_x_x as setup file of the operating software (_x_x_x = version number)
 - ip_set as the backup file of the boot file
 - Operating instructions for the BioPAT® Trace | Multi Trace as PDF file



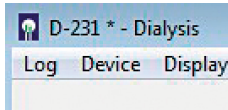
- ▶ Then change the device’s network address. It can be found in the “Network settings” menu item.

- ▶ In order to do this, you will need to log in with the password **TuSh1Vf5Ja**.

NOTICE

In most cases, the factory-set IP address will not be compatible with company networks. Have a suitable IP address assigned by your network administrator; otherwise the device may not be able to connect with the operating software.

Networking	
INTERFACE	
address	<input type="text" value="192.168.120.231"/>
netmask	<input type="text" value="255.255.255.0"/>
gateway	<input type="text" value="192.168.120.1"/>
RESOLV.CONF	
search	<input type="text" value="trace-bs.local"/>
nameserver	<input type="text" value="192.168.120.10"/>
<input type="text" value="send_data"/>	



If you would like to work via a LAN or intranet, the factory-set network address or the network name may not be compatible with your network.

- ▶ If this is the case, please contact your network administrator and change the BioPAT® Trace | Multi Trace's IP address in the form field.
- ▷ After the network address has been changed, the BioPAT® Trace | Multi Trace can only be accessed via this new IP address. The last 3 characters of the IP address (here 231) represent the number of the device that is displayed in the user interface.

6.4.1 Troubleshooting

If you have entered different or incorrect IP addresses on the device website and during the installation, it will not be possible to establish a connection. The following options are available:

- ▶ First, attempt to produce a connection to the internal device website via the standard IP address 192.168.120.231 using a direct PC connection and a cross-over Ethernet cable.
- ▶ If this is not possible, a change to a known IP address must be forced using the boot file. First switch the device off. Then copy the boot file “ip_set” onto a USB stick and connect it to the rear of the device. Then switch the device on. Once the boot process is complete, the device will now have the new IP address 192.168.120.232.
- ▶ Navigate to the new IP address 192.168.120.232 in your web browser.
- ▷ After the device has been connected in this manner, you can set the network address via your web browser as described above.

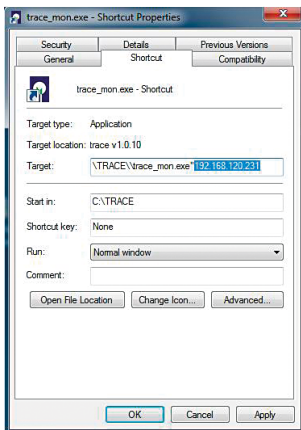
6.5 Installing the Software

- ▶ Run the file “setup_tracemon” to install the operating software on your computer. When doing so, you will be asked to enter the new IP address.
- ▶ In order to do this, overwrite the suggested factory-set IP address. If you have not changed the IP address then use “Next” to confirm.
- ▶ During the installation you will be asked to specify the storage locations for the program and the subsequent measurement data.
- ▷ Next, the software will start the device with the specified IP address and automatically connect to the BioPAT® Trace | Multi Trace.

6.6 Changing the Start Settings

If you changed the IP address at a later time, you should also update the startup settings in the program shortcut of the startup icon so that the device will continue to connect automatically. To do so, complete the following steps:

- ▶ Switch the screen view to your desktop.
- ▶ Click on the start icon with the right mouse button.
- ▶ Select the menu item “Properties”.
- ▶ Enter your new IP address at the place indicated in the figure.



- ▶ Confirm the change with “Apply” and then “OK”

6.7 Active Access Restriction | Log in

To activate the access restriction – by means of which the menu functions are assigned to the various responsible operators – the access privileges can be adjusted using the Login start command “-L”.

The device opens then when you first start the administrator mode. The administrator then creates other operators and gives them the access rights (group 0-2). For all subsequent starts initially only the lowest access level (group 0) is active and the respective operator must authorize himself using the menu item “Log in” (see Chapter “8.3.10 Menu Item “Log in””, page 105).

6.8 Assignment of Analog Outputs



The measured values remain available for further use as analog signals at the ports analog 1 and analog 2. When using the ethanol | methanol method, the signal is output on analog 1. The output analog 3 is not assigned.

Measuring channel		Measuring channel	
Pin	1 [glucose]	Pin	2 [lactate]
1	0 (2) - 10 V	1	0 (2) - 10 V
2	GND	2	GND
3	0 (4) - 20 mA	3	0 (4) - 20 mA

(Measurement channel Pin 3 is not assigned).

In the “Parameters” menu (8.3.7.1), the output format can be switched between 0 - 10 V and 2 - 10 V or 0 - 20 mA and 4 - 20 mA. The output areas of the analog outputs are also configured here.

The output areas of the analog outputs can be configured as described in Chapter “8.3.7.1 Parameters”, page 90.

The signal at the analog output is changed whenever there is a new measurement value and remains unchanged unless:

- there is a new measurement value,
- the device is rebooted,
- “new tubing set” is activated.

6.9 Checklist for Starting-up Operation

Work step	Function	Chapter	Page	Status
0	Install PC software (only on initial setup)	6.5	66	
	Access website of the device	6.4	63ff	
	Download installation program		63ff	
	Start installation program	6.5	66	
	Adjust IP address and subnet screen	6.4	63	
1	Prepare probes	6.2	56ff	
	Install membrane		57ff	
	Install probe in reactor (and sterilize, if necessary)		59ff	
2	Prepare transport buffer and waste container	6.2 and 8.3.9	56 100	
	Buffer solution (ready to use) or dilute salt mixture and prepare in bottles	6.1.5	54	
	Prepare waste container with disinfecting agent, if necessary	8.5.4	112	
3	Setup Device Tubing	6.1.2	41ff	
	Place conductivity cell into recess and affix with knurled nut	6.2.5	61	
	Attach diffusion module	6.1.2	41	
	Insert drip counters		41	
	For dialysis only: Connect three-way stopcock (optional)	6.1.3	46	

Work step	Function	Chapter	Page	Status
4	Connect inlet and outlet lines	6.1.2	41	
	Connect tubing to transport buffer		41	
	Connect tubing to calibration solutions		41	
	Connect waste line to waste container		41	
5	Switch on device	7	73	
	Switch on On/off switch on rear of the device	4.1	26	
	Wait for internal computer to boot (white LED turns red)	6.4	63	
6	Start PC software	8.1	78	
	Double-click "Start" icon on desktop (trace_mon)	8	75	
	Wait for automatic connection setup or connect device		75	
	Start with selecting "new tubing set"	6.1.5	54	
7	Set operating parameters ("Settings" menu)	8.3.7	89	
	Basic Settings		89	
	Set measurement period	8.3.7	89	
	Enter calibration standard values	8.5.1	110	
8	Connect sampling probe	4.3	33	
	Connect dialysis probe to tubing	6.2.5	61	

Work step	Function	Chapter	Page	Status
9	Starting the measurements	8.1.1	78	
	Confirm "Fill" button and wait for calibration	8	75ff	
	Alternatively: Activate the "Start autom." option – device starts measurement automatically after filling and calibration		75ff	
10	For methanol ethanol only: Connect enzyme reactor	6.1.5	54	
	Einbau des Enzymreaktor in das Schlauchset		54ff	
11	Check measuring chamber	4.3 and 6.1.2	33 41	
	Check measuring chamber for absence of air bubbles rinse if necessary		33 41	
12	For dialysis only: Perform reference measurement	8.2.1	82ff	
	Perform reference measurement with external sample or known measurement value		82ff	
13	Optional: Connect analog output to process control system	8.3.7	89	
	Connect analog output (0-10 V, 4-20 mA or 0-20 mA)		89	

7 Startup

7.1 Startup

Before every startup, check the BioPAT® Trace | Multi Trace to make sure that all modules installed on the device are functional.

The following conditions must be checked scrupulously on the device:

- Are the two original pump cassettes installed properly?
- Is the tubing set connected completely and properly?
- Is the enzyme reactor installed in the methanol | ethanol operating mode?
- Is there a conductivity cell in the holder?
- Are sufficient volumes of transport buffer and the appropriate calibration solutions ready at hand?

If these checks are passed, you can start the device. In Chapter “8 Operation”, page 75 describes in detail the operating software functions necessary to start the device. In principle, the following procedure must be maintained:

- ▶ In the “Basic Settings” menu, select the measuring method (see Chapter “8.3.7.3 Basic Settings”, page 95).
- ▶ In the “Parameter” menu, configure the calibration standards and measuring and calibration frequency you want to use (see Chapter “8.3.7.1 Parameters”, page 90).
- ▶ After you have connected all tubing and sampling lines, start the filling routine (see Chapter “8 Operation”, page 75).
- ▶ After priming, the device starts with the measurements automatically (if the autostart option was activated) or switches to the “In standby” mode.
- ▶ If you are working with the dialysis method, you must carry out a reference measurement at the right time in order to obtain conclusive results.
- ▶ Proceed as described in Chapter “8 Operation”, page 75.

7.2 Decommissioning

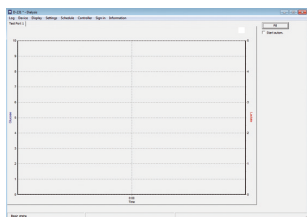
- ▶ The device can be switched off at any time by pressing the On/off switch. The calibration and reference values last stored remain in memory for the next start (see Chapter “8.3.1.1 Connect”, page 84).


If the measuring instrument is not operated for two days (e.g. over the weekend), the buffer-filled system may remain standing.

- ▶ In the event that the device will be out of use for longer periods of time, the tubing set must be replaced before re-starting operations.
- ▷ For decommissioning in the methanol | ethanol operating mode, the enzyme reactor must be removed and stored at a cool temperature (see Chapter “6.2 Connecting the Dialysis Probe”, page 56).

8 Operation

Switch on the BioPAT® Trace | Multi Trace at the On/off switch located on the rear panel and run it on an external PC.



You will find the start icon on the PC's desktop . Double-clicking it will start the application. The user interface shown in the adjacent figure appears.

The device number appears in the header. In this case: 231*. The asterisk indicates that a connection exists between the control PC and the BioPAT® Trace | Multi Trace. If there is no connection to the measuring instrument, it must be established (see Chapter “6.1.1 Device Setup”, page 39).

The measurement display can be modified at any time during operation. Press the right mouse button to display the input field for configuring the view.

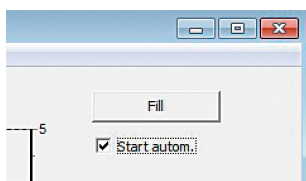
Here, you can configure dot sizes, dot colors and the axis scales.

The default settings are the color blue for glucose and red for lactate. A green dot color is preset for methanol or ethanol.

The device status is displayed under the measurement window. At the beginning, the device is in default mode. To the right of the measuring window there is an area for context-based selection buttons.

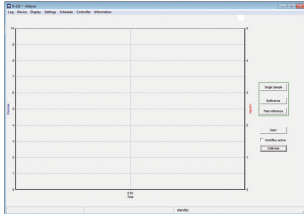
Every time the device restarts, completely fill the tubing set and run several preliminary measurements to render the sensor system stable before initiating the calibration routine. This process is called priming; alongside filling and preliminary measurements, it also includes an initial calibration of the system. After priming, the device switches to “In standby” mode. Depending on the measuring method selected, the entire process takes between 40 and 60 minutes.

- ▶ Before you start up your system with autostart, make sure that the tubing set is properly connected to the sampling outlet on the bioreactor.



Another option is to configure the BioPAT® Trace | Multi Trace to start with the measurements independently after the priming routine. If a check mark is placed next to “Start autom.”, the device will automatically initiate the measurement after performing the initial calibration routine.

If the “Fill” button is pressed, the device starts filling and the “Fill” button disappears.



When the system is ready to run the measurement, the status message "In standby" in the lower right will appear. When the priming is completed, a panel for context based selection buttons is available.

In dialysis mode, a third button labeled "Reference" appears to the right of the measuring window. If the "Reference" button is activated, the device starts with a reference measurement to calibrate the attached dialysis probe.

8.1 Basic Commands

8.1.1 Start

The “Start” command starts the measurements of the analyser. The command can either be triggered directly by pressing the “Start” button or by a corresponding entry in the schedule (see Chapter “8.3.8.1 Events”, page 97). In this case, the command will only be executed when the specified time is reached.

8.1.2 Standby

The command “Standby” stops the current action of the device. The command can either be triggered directly by pressing the “Standby” button or by a corresponding entry in the schedule (see Chapter “8.3.8.1 Events”, page 97). In this case, the command will only be executed when the specified time is reached.

8.1.3 Calibrate

The command “Calibrate” triggers a calibration. There are different options to start a calibration:

- By clicking on the “Calibrate” button
- Autocalibration (based on a number of measurements performed in between, see Chapter “8.3.7.1 Parameters”, page 90)
- By a corresponding entry in the event list (see Chapter “8.3.8.1 Events”, page 97). In this case the command will be proceeded when the respective time is reached.
- By a corresponding entry in the workflow (see Chapter “8.3.8.2 Workflow”, page 98).

8.1.4 Single Sample

This function can be used for a spontaneous unscheduled single point measurement at a selected port (Multi Trace). This function is especially useful during long intervals, when for instance an actual value of a certain reactor is wanted. By this option, the running program does not need to be interrupted and the planned intervals remain unchanged.

The "Single Sample" command is only triggered, if there is enough available time for the measurement.

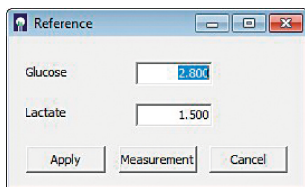
8.2 Reference (for Dialysis only)

A reference measurement must be performed to compare the permeability of the two diffusion membranes in the diffusion cell (which is only allowed contact with calibration solutions) and in the dialysis probe (which only has contact with the reactor contents). The relationship of the two membranes to one another is expressed by a reference factor used for calculating the measured values.

You can conduct a reference measurement whenever you want, in particular when changes occur in the process that may influence the diffusion speed (e.g. as a result of temperature changes).

Reference measurements can also be carried out at regular intervals, e.g. when samples are taken for external accompanying analysis. The measured value is only corrected with a new reference factor when the new reference value was entered and confirmed by pressing "Apply". Up to then, measured values that were determined in the meantime are always calculated and displayed with the currently valid reference factor.

The device starts with a reference factor of 1.0. If the two membranes were permeable to the same degree, then this value would be correct. In practice, however, experience has shown that manufacturing-related and ambient conditions affecting the probe can lead to discrepancies.



The comparison is activated as follows: After clicking the “Reference” button, the device determines the concentration values in the reactor and suggests the corresponding values. Now, using a reference sample, you must check whether these values are correct and correct them accordingly. If you already know the concentration values (e.g. from the initial weight), you can enter these values directly. Alternatively, you can determine the concentrations in the reactor by means of an external sample using an off-line analyzer.

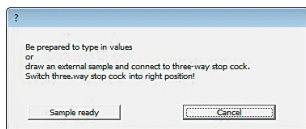
Confirm the prompted measured values with “Apply”. If you want to make changes, enter the new values into the entry field (with decimal point) and confirm with “Apply”

With the “Reject” function, you can cancel the procedure and move back to the starting position.

As long as the input window is open and no new reference values were entered and confirmed, the BioPAT® Trace | Multi Trace continues working with the current valid reference factor.

If determining a reference measurement using an external analyzer is going to take longer and the medium concentration is expected to change in the meantime, you should wait for the reference measurement and only then take a sample for external analysis. When the input window for the reference values appears, the device has stored the currently measured concentration to compare it with subsequently entered values and determine the new reference factor.

If no off-line analyzer is available for determining reference values, you can also measure the sample on the BioPAT® Trace | Multi Trace.



- ▶ To do so, select the “Measurement” function and follow the further instructions prompted by the menu navigation.



- ▶ Draw a cell-free sample from the bioreactor (at least 5 mL) into a 5 mL syringe and connect it to the three-way stopcock. Check that the three-way stopcock is in the correct position: The arrows on the stopcock must point towards the green tube marking and the syringe.

NOTICE

Only use the “Sample ready” function if you are sure that the filled syringe is connected and the three-way stopcock is set to the proper position. Otherwise erroneous measurements may result!

“Sample ready” is activated to measure the external sample. Three measurements are taken and their average value displayed as a result. As described above, you can then accept, correct or discard the measurements or you can perform another measurement on an external sample.

NOTICE

Make sure that the three-way stopcock is set as displayed. Otherwise all following calibrations will be erroneous and the resulting measurements incorrect!



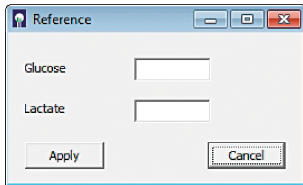
Before you continue working, make sure that the three-way valve was set back to the initial position after removing the external sample, so that you can correctly measure Standard 1 during the next calibration.

8.2.1 Fast Reference (Only for Dialysis)

In use of the dialysis method it is necessary to perform a referencing in order to match the permeability of the membranes within the reactor and outside in the diffusion cell, and to compensate for temperature differences between the two membranes. A reliable determination of the reference factor is therefore very important for the quality of the measurement results.

Normally, a measurement is triggered by the "Reference" option and the measurement result is communicated. The user can then confirm the measured values (reference factor does not change), correct the value or perform the analysis of an external sample. For processes with slow concentration changes (e.g. cell cultures) and already known concentration values in the medium, this procedure can be accelerated by using the "Fast Reference".

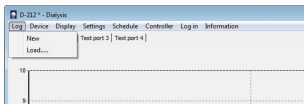
The “Fast Reference” option allows a faster implementation of the reference measurement, in which the known concentration values of the medium can be entered before the actual reference measurement. After pressing the “Fast Reference” button, the input window appears.



Now enter the corresponding concentration values and confirm with “Apply”. The device will then begin with the reference measurement and when finished automatically returns to the state it was in when the “Fast Reference” button was triggered. “Reject” will cancel the operation and return to the starting position.

As long as the input window is open and no new reference values have been registered and confirmed, the BioPAT® Trace | Multi Trace Control still works with the current reference factor.

8.3 Menu Bar



The measured data are recorded as text in a bpd file and saved in the default subdirectory “Data”.

In addition to the date and time the respective data for temperature at the measurement cell, glucose reading and lactate reading will be logged. If the controller is active, the respective feed rate (1 = 100%) is recorded. If the device is in the Multi Trace mode, the port for which the values apply is logged as well.

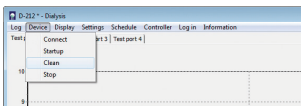
If you want to create a new file, select the menu item “New”.

If you want to load and display measured data from an old file, select the menu item “Load...”.

The saved files can be easily processed, e.g. in Excel. For notes on importing see Chapter “8.5.5 Import Data to Excel”, page 113.

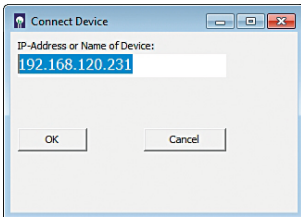
8.3.1 Menu Item “Device”


The user can choose from four sub-items: “Connect”, “Startup”, “Cleaning” and “Stop”.



8.3.1.1 Connect

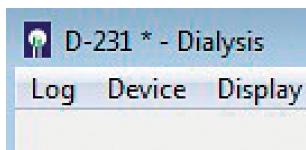
By selecting the “Connect” menu sub-item, you have the option to connect a new BioPAT® Trace | Multi Trace. To do so, you must enter the IP address of the device.



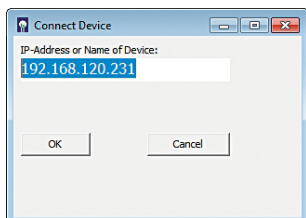
If you have started the software by clicking on the start icon  on the desktop, the standard **IP address 192.168.120.231** will already be displayed.

If your device has been assigned a different IP address (e.g. by your network administrator), then enter that address here.

If you would also like to be prompted with this new address automatically in the future, proceed as described in Section “6.6 Changing the Start Settings”, page 67.



Once you have entered the correct IP address and the device is acknowledged by the network, it will connect to the control PC. An asterisk appears in the view bar behind the name or number of the device.



Alternatively, you can start a search for new devices on the network by clicking on the button “Who is out there?”. The results will be displayed after a little while. You can highlight the desired device and select it with “Ok”.

If no device can be found, an error message appears. Try to enter the IP address directly or contact your network administrator.

8.3.1.2 Startup

If the tubing set has been replaced, you must go to the menu item “Startup”. “Startup” deactivates all previous calibration and reference data and requires that the tubing system is re-primed.

8.3.1.3 Cleaning

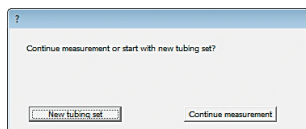
For longer waiting times the cleaning mode should be used. For this purpose, all of the lines of standards and transport buffer, and optionally the sample feed has to be connected to the cleaning solution (Order No. BPT0044A / BPT0044G). Then in the menu “Device” the option “Cleaning” is selected.

This option starts an automatic cleaning procedure, including a 5-fold cycling of the priming mode. Afterwards, both pumps flush the tubing set at the low flow rate of the standby mode.

For a new start, you must go to the menu item "Startup" (see Chapter "7.1 Startup", page 73). "Startup" deactivates all previous calibration and reference data and requires that the tubing system is re-primed (with transport buffer!).

8.3.1.4 "Stop"

"Stop" is used for temporary decommissioning of the system, e.g. when the monitored bioprocess is interrupted, for short operating interruptions or when connecting enzyme reactors (see Chapter "7.2 Decommissioning", page 74). Select the menu item "Stop" to interrupt the measuring routine and ready status. All pumps stop and the valves shut. In this event, the device will retain stored data on calibration and reference values.



Click the "Stop" button and the BioPAT® Trace | Multi Trace switches to standby. The program can now be closed. The device can also be switched off at the On/off switch without losing any data. When switching the device back on or re-booting the control program, the prompt screen shown in the adjacent figure appears.

By clicking on the button “Continue measurement”, the system switches directly to operational mode. No rinsing cycles or calibrations will be carried out. The former calibration values will be retained.

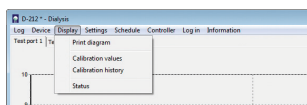
NOTICE

Do not activate “Continue measurement” unless you are certain that the same conditions prevail as before the pause (e.g. in the event of brief interruptions). Otherwise, this may result in erroneous measurements!

In the case of longer interruptions (e.g. overnight), it is recommended to carry out a calibration before continuing with further measurements.

8.3.2 Menu Item “Display”

When the menu item “Show Connection” is activated, an asterisk indicates that the control PC is communicating properly with the BioPAT® Trace | Multi Trace.

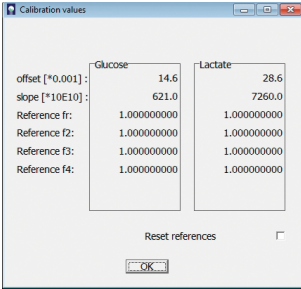


8.3.3 Print Diagram

By selecting the menu item “Print diagram”, the currently displayed diagram is printed out on an interfaced printer.

8.3.4 Calibration Values

When the menu item “Calibration values” is selected, a window opens that shows the current calibration values.

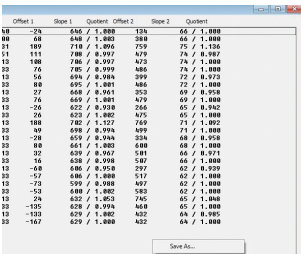


The axis intercepts (a0), calibration gradient (a1) and the reference factors (Reference fr) are displayed.

Activating the option “Reset reference” resets the reference factors to the factor 1.0 (only in dialysis mode).

8.3.5 Calibration History

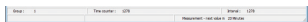
When the menu item “Calibration history” is selected, a window opens that shows the current calibration history.



By clicking the right mouse button, the calibration history can be saved as txt.-file.

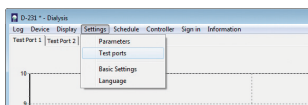
8.3.6 Status

When the menu item “Status” is selected, an information bar appears at the bottom edge of the screen that displays further information on the current status in three fields. This function is of particular interest for long intervals or group measurements.



The number of pending group measurements (or preliminary measurements) is displayed in field 1. In field 2, a current time count counts down the time until the end of the current action (in seconds). The remaining time in the interval of the measurement period is displayed in field 3 (in seconds).

8.3.7 Settings

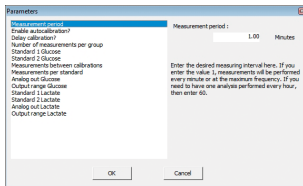


In the “Settings” menu options, you can configure the most important device settings for operation and communication:

- Parameters
Allow the user-specific configuration of measurement frequency, calibration frequency, concentration of the calibration standards etc.
- Test ports (Multi Trace)
Here, up to four test ports are activated. This is the case when using the BioPAT® Multi Trace and individual reactors have not yet been put into operation or should not be used. At least one test port must remain activated.
- Basic Settings
Here you can configure the device for the selected sampling method. Likewise, the user can enter information about the device and the project that later appear on the data output.
- Language
Here you can change the menu guidance language.

8.3.7.1 Parameters

All relevant parameters for measuring operations can be configured in the “Parameters” sub-item.



Measurement Period

- ▶ Enter the desired measuring interval here. When the value 1 is entered, measurement is performed every minute or at the maximum frequency. If one analysis should be performed every hour, then enter 60.
- ▷ The frequency can be changed at any time and becomes immediately effective by clicking “Ok”.

Autocalibration

- ▷ Select yes/no. If you allow autocalibration, recalibration takes place automatically after every x number of measurements (see: measurements between calibrations).

Delay Calibration

- ▶ Normally, a calibration is performed after a certain number of measurements. Longer interruptions (e.g. measurements every 6 hours for cell cultivations) can lead to very long periods expiring between the most recent calibration and the next measurement. Activating the Delay calibration function schedules the calibration to be performed shortly before the next measurement. This way, an up-to-date calibration is available.

Number of Measured Values in Groups

- ▶ When using larger measuring intervals, it can make sense to run several measurements with maximum frequency in quick sequence at a given point of time. This way, for example, a group of 10 measurements may be carried out every 6 hours. The number of measured values in the group can be set here. No average is calculated. Every measured value is displayed immediately after measurement. If the time required for the performance of the group is longer than the measurement period, then the next group starts with the new measuring interval.

For the determination of the number of measurements between calibrations, every measurement in a group is taken into account. This must be given due consideration when selecting the calibration settings!

Standard Levels (Glucose and Lactate)

- ▶ Here, you indicate the concentrations of the calibration standard solutions that have been selected and hooked-up for your application.

Measurements Between calibrations

- ▶ Here, you can enter how many measurements should be performed before an automatic calibration is run. This feature only works, however, if the autocalibration function has been allowed.

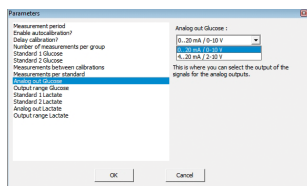
By pressing the “Calibration” button, you can initiate a calibration at any time. Any changes made to the automatic calibration data during running operations are not valid until the next calibration.

Measurements Per Standard

- ▶ This function is used to set the number of measurements per calibration standard. One minute is required for each measurement of a calibration value. That means that a normal calibration with 3 measurements per standard will take 6 minutes (including rinsing times for changing the medium).

Analog Out Glucose | Analog Out Lactate

- ▶ Here, the output of the signals on the analog outputs can be selected between 0–20 mA and 4–20 mA, and between 0–10 V and 2–10 V, respectively.



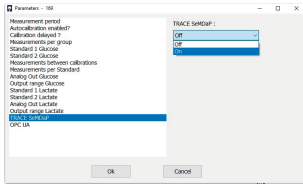
Output Range Glucose | Lactate

- ▶ Here, the output ranges of the analog outputs are set. Proceed as follows: The upper limit of the measuring range is always indicated. The lower limit is preset to 0.

For a required measurement range of e.g. 0–10 g/L glucose, glucose is set to 10 (upper limit) in the output range.

When selecting the voltage range of 0–10 V, then 10 volts are equal to 10 g/L glucose.

Data Output via the Serial Interface



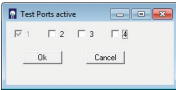
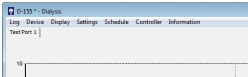
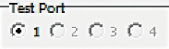
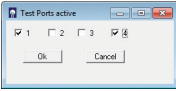
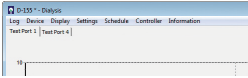
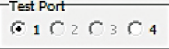
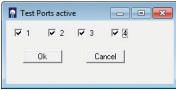
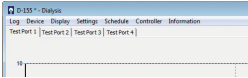
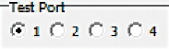
The TRACE SeMDaP is a protocol for outputting data via a serial interface. This serial data protocol contains data of variable length and content. Outputting of the serial data protocol is switched off at the factory.

Output of the serial data protocol is activated by the following tasks:

- ▶ Select [TRACE SeMDaP] for the parameter
- ▶ Select [ON] for [TRACE SeMDaP]
- ▶ For further details on protocol, please contact Sartorius Service.

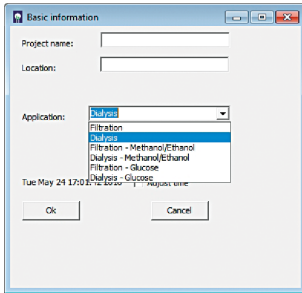
8.3.7.2 Test Ports

For the BioPAT® Multi Trace version the unit has 4 different test ports (bioreactor probes 1–4). These are controlled by the selection valves on the side of the device. In the case that not all four measuring probes are intended to be used, the respective activation of each port can be specified using the tag input window. The corresponding ports are then differently displayed in the scheduler and in the selection bar of the user interface. The setting also has an effect on the number of displayed graphics.

Test ports	Control panel	Graphics displayed	Schedule
	<p>selected: 1</p> <p><input checked="" type="radio"/> Probe 1</p> <p><input type="radio"/> Probe 2</p> <p><input type="radio"/> Probe 3</p> <p><input type="radio"/> Probe 4</p> <p>Only probe 1 active</p>		<p>Test Port</p>  <p>Only probe 1 active</p>
	<p>selected: 1</p> <p><input checked="" type="radio"/> Probe 1</p> <p><input type="radio"/> Probe 2</p> <p><input type="radio"/> Probe 3</p> <p><input type="radio"/> Probe 4</p> <p>Probe 1 and 4 selectable</p>		<p>Test Port</p>  <p>Probe 1 and 4 selectable</p>
	<p>selected: 1</p> <p><input checked="" type="radio"/> Probe 1</p> <p><input type="radio"/> Probe 2</p> <p><input type="radio"/> Probe 3</p> <p><input type="radio"/> Probe 4</p> <p>All probes selectable</p>		<p>Test Port</p>  <p>All probes selectable</p>

BioPAT® Trace has only one test port, therefore the checkboxes are unchecked by default or are not displayed.

8.3.7.3 Basic Settings



In the “Basic Settings” menu item, you can enter data that is important for carrying out and documenting the measurements.

Before running the measurement, you can enter the project name and device location. This information will later be displayed in the header enabling a better assignment of data to the process, especially if several devices are in operation at the same time.

You can either select between the dialysis for the determination of glucose, lactate and ethanol, as and the dialysis methanol | ethanol.

If glucose as sole parameter should be measured the method dialysis-glucose can be selected. The lactate measurement channel will be disabled in this application.

⚠ CAUTION

Before working with a new tubing set, always check to make sure that the right application has been selected! Otherwise, this can result in erroneous measurements or destruction of the tubing set!

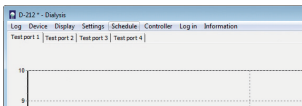
By placing a check mark next to “Set time”, you can compare the system times on your PC and the BioPAT® Trace | Multi Trace. It will then use the PC’s system time.

8.3.7.4 Language



You can select your preferred language under the “Language” menu item. German, English, French, Spanish, Italian and Chinese are available.

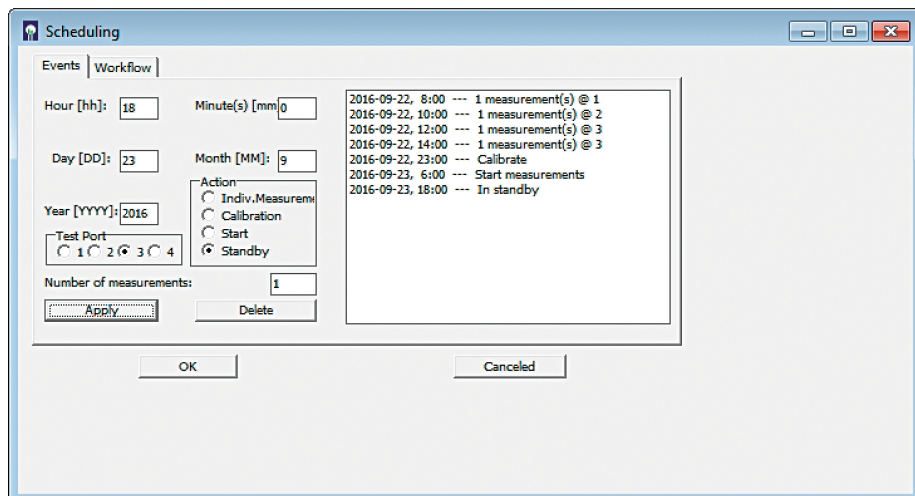
8.3.8 Menu “Schedule”



Using the menu “Schedule”, the device can be individually programmed in its temporal actions. The schedule is divided into the sections “Events” and “Workflow”. The schedule can be viewed at any time.

- ▶ **“Events”** are certain actions at fixed time points.
- ▶ **“Workflow”** is a cyclically recurring sequence of individual actions that are carried out until the device is placed in standby or completely stopped.

8.3.8.1 Events



In the submenu “Events”, individual actions are entered at certain times. For every action (one shot measurement, calibration, start, standby) one or more time points can be determined. These events are always handled in priority to other actions (from the schedule or the parameter list). In the event that the scheduled time of an action is conflicting with an action of the schedule or in the parameter list, it will wait until this is finished.

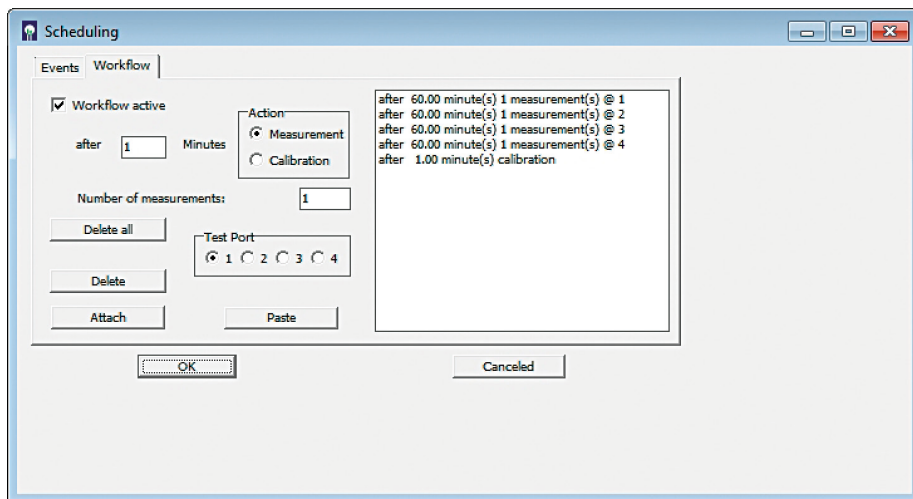
It is possible to switch the device at a specific time to start (“Start”) and later again in standby (“In standby”). During the measurements either the workflow is executed (if enabled, see below “8.3.8.2 Workflow”, page 98) or the measurement frequency and possibly edited autocalibrations according to the parameter list.

If several test ports (BioPAT® Multi Trace) are present and enabled, the respective measurements and the desired test port (1–4) can be assigned. The number of individual measurements (corresponding to “Group” in the parameter list) at the specified time is set via the input field “Number of Measurements”.

By pressing the “Apply” button, the settings selected are transferred to the right list box and arranged chronologically. The entry can be marked by clicking on a line and removed from the list again by pressing “Delete”. Double entries will be ignored, or at the same time entries only the command above is processed.

Pressing the “OK” button, all records are saved and activated. Press “Canceled” to discard the changes. After a scheduled event was carried out, the entry is removed from the list and does not appear at a later opening of the schedule again. Thus, only the pending events are displayed.

8.3.8.2 Workflow



The sequence of actions is created by registration of the time interval, the desired action (measurement or calibration), the number of individual measurements and the corresponding test port. The time interval between the individual actions is entered in the field “after ... Minute(s)”. If “1” is entered, the next action is performed after a minimum time interval (rinsing of the probe). By choosing larger intervals, the measurement frequency is reduced accordingly.

The workflow is enabled by a check mark "Workflow active" in the selection box. If this box is not checked, the measurement processes in the parameter list are processed instead.

The selected action for the first time interval in line 1 is active only during the second pass of the list. This ensures that the processing of the list begins right after the start. On the second pass the selected first time interval in line 1 applies.

The number of individual measurements (corresponding to "Group" in the parameter list) is set via the input field "Number of Measurements". If several test ports (BioPAT® Multi Trace) are present and enabled, the respective measurements and the desired test port (1-4) can be assigned.

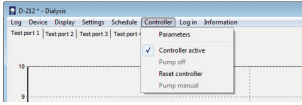
By pressing the "Attach" button the selected settings are transferred to the right list box and entered in the bottom row. If an action has to be placed between two existing entries, the lower line must be marked. Then, pressing the "Paste" button inserts the selected setting. The entry can be marked by clicking on a line and removed from the list again by pressing "Delete".

Pressing the "Clear" button all entries will be deleted from the list box.

Pressing the "OK" button, all records are saved and activated. Press "Canceled" to discard the changes.

The workflow is executed cyclically until the unit is stopped or put into standby.

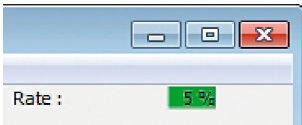
8.3.9 Menu “Controller”



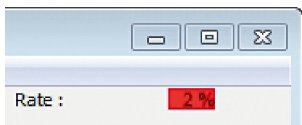
The integrated controllers (PID Controller and Minimum-Maximum Controller) can be configured and operated in the menu “Controller”. The controllers can directly drive external pumps (via the serial interface or analog output 3 on the back of the device) as feeding pumps.

For BioPAT® Multi Trace the controller is also available, but only in single-channel operation. If several test ports are measured, the controller may not be used, since the respective last signal (no matter what test port) is used for the controller as an input variable, thus resulting faulty feedings.

On the display a box with the indication of the current feed rate (0–100%) of the feeding pump appears when the pump is active.

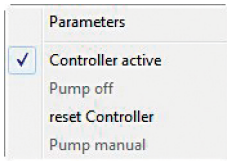


A green background indicates an active controller.

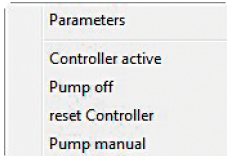


The background turns red when the controller is inactive.

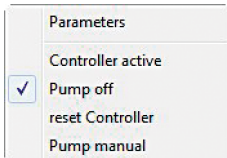
The respective feed rates are recorded in the bpd-file (see Chapter 8.2.1).



If “Controller active” is selected, the built-in device controller is switched on and the feeding pump is activated. While the controller is active, the pump cannot be switched off or operated manually.

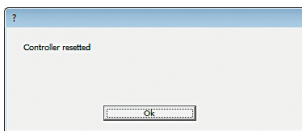


To disable the controller, the check mark must be clicked. In this state, the controller is inactive, but the feeding pump is still running at the rate specified by the last controller on.



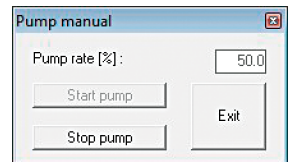
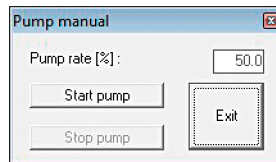
With “Pump off” the feeding pump is switched off manually. You can then turn on again only by the re-activation of the controller.

In order to avoid a too high influence of the integral gain on the PID Controller at the beginning, the integral gain can be reset to the preset value by “Reset controller” at any time.

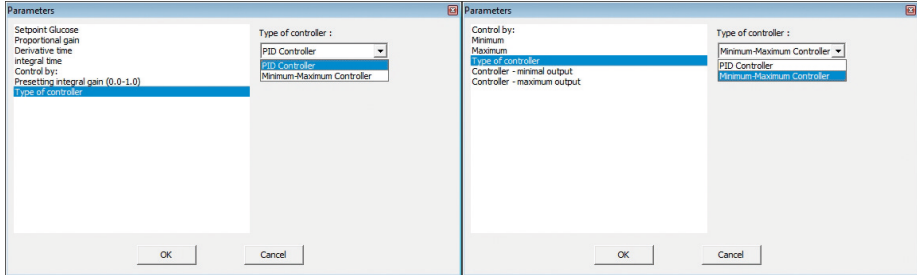


This command must then be confirmed with “Ok”. This function has no effect on the Minimum-Maximum Controller.

“Pump manually” is a function that can be used for filling the feeding tube. In this operating window, the pump rate can be adjusted and the pump is started or stopped. By “Exit” or close the window, the pump is automatically stopped.

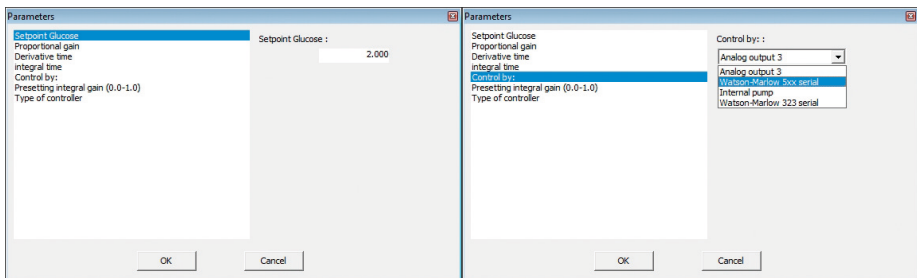


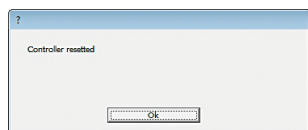
The respective type of control can be selected between PID Controller and Minimum-Maximum Controller in the sub menu “Parameters” under “Type of controller”.



8.3.9.1 PID Controller

After selecting the type of controller to PID Controller in the submenu “Parameters” the “Set-point Glucose” is registered for the control. Moreover, here the control parameters “proportional gain”, “derivative time” and “integral time” can be entered. In the option “control by” it can be selected if the control acts to the integrated pump or an external pump via the serial interface or analog output 3.





In order to avoid a too high influence of the integral gain on the controller at the beginning, the integral gain can be reset to the preset value by “Reset controller” at any time. This command must then be confirmed with “Ok”.

⚠ CAUTION

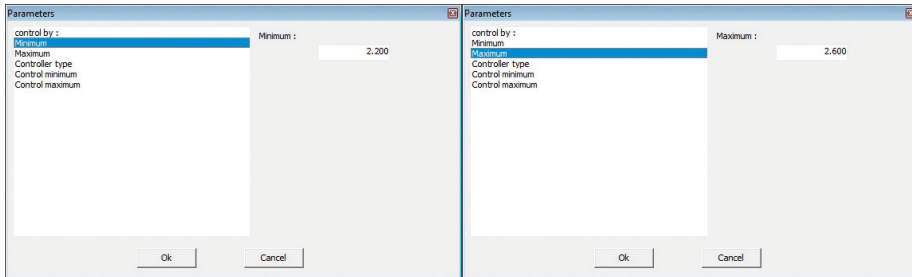
Always reset the controller after filling the feeding line or with unknown history to avoid overdose!

Recommended start settings for the PID Controller:

- ▶ For microbial cultures the following settings can be recommended:
 $P = 0.1-0.5$ (proportional gain). Depending on the concentration of the feeding solution, max. pump speed, diameter of the tubing, reactor volume, etc.,
 $I = 45$ min (integral time),
 $D = 2$ min (derivative time).
- ▶ For cell cultures the following settings can be recommended:
 $P = 0.01-0.1$ (proportional gain). Depending on the concentration of the feeding solution, max. pump speed, diameter of the tubing, reactor volume, etc.,
 $I = 60$ min (integral time),
 $D = 1$ min (derivative time).
- ▶ In most cases adjustment of the P-value and you can let the I-value and D-value at the recommended level.
- ▶ If the feeding is too strong reduce the P-value or if it is not enough increase the P-value.

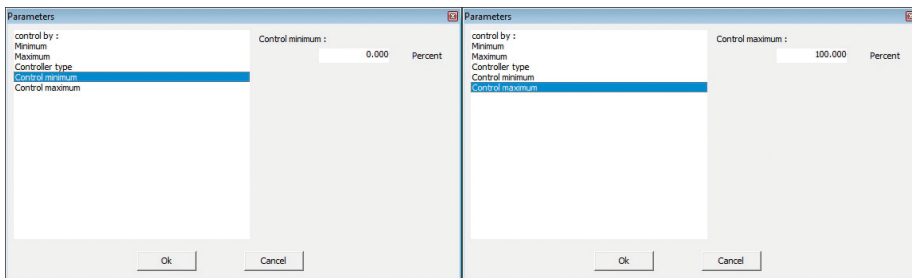
8.3.9.2 Minimum-Maximum Controller

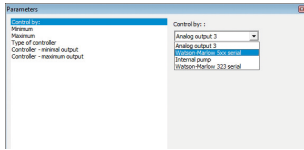
After selecting the type of controller to Minimum-Maximum Controller in the submenu "Parameters" the "Minimum" and "Maximum" limits are registered for the control.



The Minimum-Maximum Controller switches the feeding pump if it is above or below the limits respectively to the minimum flow rate, or maximum flow rate.

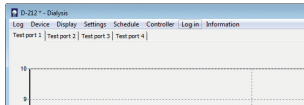
The minimum delivery rate and the maximum flow rate of the pump can be specified with the setting "controller - minimal output" and "controller - maximal output". The default settings are preset to 0% and 100%.





In the option “Control by” it can be selected if the control acts to the integrated pump or an external pump via the serial interface or analog output 3.

8.3.10 Menu Item “Log in”



This menu can set up a server management that allows different access rights depending on the responsibility of the user.

This menu item is only activated if the program was started with the launch setting “-L” (see Chapter “6.7 Active Access Restriction | Log in”, page 68). The device opens then when you first start the Administrator mode. The administrator then creates other operators and gives them the access rights (group 0-2). For all subsequent starts, initially only the lowest access level (group 0) is active and the respective operator must authorize himself using the menu item “Log in”.

The following table “8.1 Menu functions” shows the assignment of the preset access rights to the respective groups.

8.3.11 Menu Item „Information”

After selecting the menu item „information”, the following data will be displayed:

- The Current version-number of the software.
- The Current version-number of the control software trace_mon.
- The Serial number of the device.

Menue function	Administrator	Responsible	Operator	All
Name	admin			
Password	trace			
Group	6	2	1	0
Log	+	+	+	
New	+	+	+	
Open	+	+	+	
Device	+	+	+	
Connect	+	+	+	
Startup	+	+	+	
Cleaning	+	+	+	
Stop	+	+	+	
Display	+	+	+	+
Print diagram	+	+	+	+
Calibration values	+	+	+	+
Reset reference	+	+	+	
Calibration history	+	+	+	+
Status	+	+	+	+

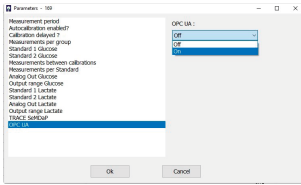
Menu function	Administrator	Responsible	Operator	All
Settings	+	+	+	+
Parameters	+	+	+	
Test ports	+	+	+	
Basic settings	+	+	+	
Language	+	+	+	+
Schedule	+	+	(view only)	(view only)
Controller	+	+		
Parameters	+	+		
Controller active	+	+		
Pump off	+	+		
Reset controller	+	+		
Pump manually	+	+		
Login	+	+	+	+
New	+			
Delete	+			
Log in	+	+	+	
Change password	+	+	+	
Information	+	+	+	+
Function keys (basic commands)	+	+	+	

8.3.12 Data Transfer via TCP/IP

The device can send data to external clients as an OPC UA server.

The OPC UA server is activated by the following tasks:

- ▶ Select [OPC UA] for the parameter
- ▶ Select [ON] for the [OPC UA]
- ▶ For further details on protocol, please contact Sartorius Service.



8.4 LED Status Messages

In order to be able to read the device status independently of the operating software on a connected PC directly on the BioPAT® Trace | Multi Trace, the various operating statuses can be read using the LEDs on the front panel. The table “8.4 Status messages of the LED display” provides an overview of the LED signals:

Status	Action	LED1 (green)	LED 2 color	LED 2 status
Startup and decommissioning				
Boot process		on	white	on
Stop		on	cyan	on
Filling		on	cyan	flashes
Cleaning		on	magenta cyan	flashes
Preliminary measurement		on	green cyan	flashes
Weighing				
Weighing	loaded	on	green	on
Weighing	unloaded	on	green	flashes
Measurement breaks				
Readiness	Readiness	on	yellow	on
Interval	standstill	on	yellow	flashes
Rinsing cycles				
Changing the medium		on	magenta	on
Rinsing probe		on	magenta	flashes

Status	Action	LED1 (green)	LED 2 color	LED 2 status
Calibrations				
Calibration	loaded	on	blue	on
Calibration	unloaded	on	blue	flashes
Reference	loaded	on	blue	on
Reference	unloaded	on	blue	flashes
Malfunctions				
No network connection		on	red	on
Initialisation error		on	black	off

8.5 Tips

8.5.1 Choosing Suitable Standard Solutions

Various combinations of calibration standard solutions are available to optimally cover the expected concentration cycles. Sartorius recommends connecting a higher concentrated standard than standard 1.

Depending on the process conditions and the objective of the cultivation, various concentration ranges have to be covered. The concentration range should principally be between the concentrations of the standard solutions in order to ensure optimal measuring accuracy. Furthermore, we recommend always choosing a standard with very low concentration in order to properly determine measured values close to zero.

For regulation, the concentrations of the calibration standards should not be too far from the regulation (set)point. For regulation at 1 g/L, for example, it is more practical to use a combination of 0.5 g/L and 4 g/L than a combination of 1 g/L and 10 g/L, for example.

8.5.2 Dialysis Mode: Referencing at the Beginning of a Fermentation

It is crucial to perform reference measurements when using the dialysis method to compare the permeability of the membranes inside the reactor and outside the diffusion cell as well as to compensate for temperature differences between both membranes. Thus, reliable determination of the reference factor is very important for the quality of the measured values.

It is therefore recommended that you determine the reference value as soon as possible at the start of the cultivation and do so in a phase without fluctuations in the concentration of the medium. For this purpose, deactivate auto-calibration and then set the measuring interval to 1. Afterwards, track some measured values. If they do not spread too widely and are as constant as possible, you can trigger reference measurement. Once the reference measurements are completed, you should wait for some measurements and check if the recalculated values meet your expectations. If necessary, you can also repeat the reference measurement. As soon as you are satisfied with the result, you can readjust the measurement parameters and start with the cultivation.

8.5.3 Dialysis Mode: Referencing at Low Lactate Concentrations

In many cases, there is a surplus in glucose and the medium is (still) lactate-free at the start of a cultivation. When performing reference measurements, the BioPAT® Trace | Multi Trace suggests a value once the reference measurement is completed. This value is usually overwritten by entering the known value (known due to the media composition or determined with an external analyzer). If the available lactate concentration is still very low, do not yet overwrite this value (the reference factor for lactate is still 1.0). During the course of the cultivation, the increase in lactate concentration can now be observed more easily. In due course, a standard reference measurement overwriting both values can be performed.

8.5.4 Avoiding Contaminations in the Tubing Set

During longer processes (over a period of weeks), biofilm can form inside the tubing set over time. This can result in the lines clogging or in highly fluctuating measured values.

This applies in particular to storage tanks with ready-for-use diluted transport solution that are repeatedly refilled with fresh solution. It is advantageous to completely replace the almost-empty container with a new container with fresh solution to avoid a transfer of microbes.

The rinsing line of the conductivity cell when removing air bubbles in the measuring chamber presents another opportunity for microbes to intrude into the tubing set. Therefore, it is imperative to use only fresh solutions and | or a sterile filter attachment for this purpose.

In some long-term cultivations, it was observed that biofilm would form in the waste line and spread into the drip chamber. In this case, it is also recommendable to regularly replace the waste containers and to prevent the waste line from being immersed in the waste. The waste containers can be further protected by adding cleaning solution (see Chapter “3.6 Consumables”, page 21).

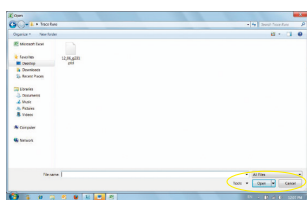
8.5.5 Import Data to Excel

The measured values of the BioPAT® Trace | Multi Trace are saved on the PC as a bpd file.

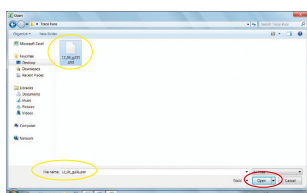
For further processing, the data can be imported into Excel.

The data import of the bpd file into Excel is carried out in seven steps:

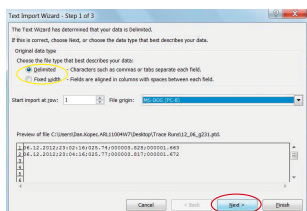
- ▶ Open the MS Excel program.
- ▶ Select “File” | “Load” and select the directory in which the bpd file is saved.
- ▶ Activate “All files” (circled in orange)

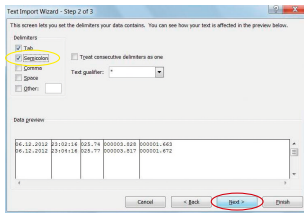


- ▶ Highlight the bpd file (circled in orange)
- Confirm this selection by pressing the “Load” button (circled in red).

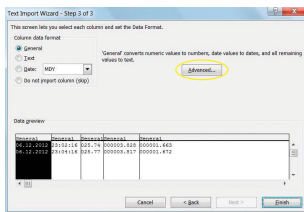


- ▶ Text conversion assistant (Step 1)
- The text conversion assistant will then start. The file is read in three steps. First, select the field “Separated” (circled in orange). This step is completed by pressing the “Skip” button (circled in red).

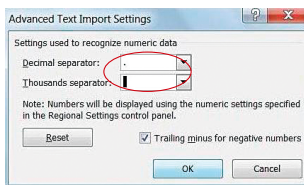




- ▶ Text conversion assistant (Step 2)
In this step, the “Semicolon” separator is selected (circled in orange) and confirmed by pressing the “Next” button (circled in red).



- ▶ Text conversion assistant (Step 3)
In this step, the “More” option is selected (circled in orange).



- ▶ Here you can select “More text import options”. A decimal point “.” is set as the decimal separator and a space as the separator for thousands (circled in red).

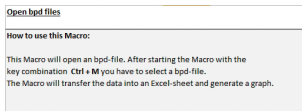
- ▶ After confirming this window by pressing “OK”, confirm step 3 of the text conversion assistant by pressing “Skip”. The data from the bpd file are now available as an Excel spreadsheet. In addition to the date and time the respective data for temperature at the measurement cell, glucose reading and lactate reading will be logged. If the controller is active, the respective feed rate (1 = 100%) is recorded. If the device is in the BioPAT® Multi Trace mode, the port for which the values apply is referred to in column 3 (the numbering starts at 0 for test port 1).

8.5.6 Data Transfer to Excel Using an Excel Macro

Alternatively to the manual procedure the data can be transferred to excel using the macro "Macro_bpd.xlsm". This macro is stored in the device's data memory and can be accessed via the internal device website (see Chapter "6.4 Loading the Software and Network Connection", page 63). This macro automatically creates a table and a graph.

Procedure:

- ▶ Copy "xx.bpd" and "Macro_bpd.xlsm" in a new folder
- ▶ Rename "xx.bpd" as "data.bpd"
- ▶ Open "Macro_bpd.xlsm"
- ▶ Start macro with "Ctrl+M"
- ▷ Creates a file with table and graph
- ▶ Stored as Data_Trace_YYYY_MM_DD.xlsm



9 Maintenance and Servicing

9.1 Cleaning

- ▶ The surface of the BioPAT® Trace | Multi Trace housing should only ever be cleaned with a wet cloth.

9.2 Decontaminating the Tubing Set

9.2.1 Contamination by Biofilm

During processes that run over long periods of time (periods of more than one week), the inside of the tubes becomes contaminated by a biofilm. This biofilm causes the tubing sets to become increasingly blocked and leads to fluctuating and inaccurate readings. This particularly applies to the storage tanks, which are constantly being filled with new, diluted “ready-to-use” transport solutions.

To prevent the transmission of microbes, the almost empty storage tanks must be completely replaced by new storage tanks with fresh solution.

During some cultivation processes that run over long periods of time, the biofilm also forms in the disposal line and the drip chamber. In this case, too, it is necessary to replace the disposal tank regularly. This prevents the disposal line from being permanently filled with “waste”.

The disposal tank can be protected by also filling it with a disinfectant solution.

9.2.2 Causes of Contamination

- Connection lines: The connection lines are neither cleaned nor disinfected prior to use.
- Transport buffer: The transport buffer is prepared in a contaminated container or filled into an already used standard container.
- Tubing set: The tubing set is used again without having been thoroughly cleaned or disinfected beforehand.
- Disposal line | disposal tank: The disposal line is filled with liquid. No disinfectant was added to the disposal tank. The disposal tank has not been closed.
- Probe sterilization: The probe has not been properly sterilized or contaminated with a rinse solution.

9.2.3 Decontaminating Connection Lines

- ▶ Rinse the connection lines with a cleaning solution.
- ▶ Rinse each connection line with 5 ml isopropanol prior to installation. More extended contact should be avoided, as long-term contact with isopropanol may damage the adhesions inside the connecting piece.

9.2.4 Decontaminating the Transport Buffer

- ▶ Only use cleaned transport containers for transportation.
- ▶ Do not refill the transport container with fresh transport buffer, but use a new | cleaned transport container.
- ▶ Filter the buffer under sterile conditions (in accordance with GMP).

9.2.5 Decontaminating the Tubing Set

- ▶ Clean the tubing set with a cleaning solution (BPT0044A/BPT0044G).

9.2.6 Disposal Line | Disposal Tank

- ▶ Add some KOH or NaOH cleaning tablets to the disposal container. Use of a chemical disinfectant is also possible.
- ▶ Seal the disposal container airtight with “Parafilm” or a similar sealing film.

9.2.7 Probe Sterilization

- ▶ Check whether there has been any contamination after sterilization.
- ▶ Avoid rinsing the probes with a non-sterile liquid after sterilization.

9.3 Maintenance and Servicing Activities

9.3.1 Device

If a measurement series is interrupted for a short time (maximum of two days), the system can continue to be operated without replacing the tubing set.

- ▶ If the BioPAT® Trace | Multi Trace is to be decommissioned for longer periods of time (more than 2 days), the tubing set should be replaced upon recommissioning (see Chapter “6.1 Setup”, page 39).

9.3.2 Tubing Set

Clogged or damaged tubing sets should be replaced with new ones.



- ▶ Before every startup of the system, check all connections made with Luer lock or UNF fittings for any leaks and tighten them as necessary.

10 Malfunctions

If the BioPAT® Trace | Multi Trace will not start or malfunctions occur during operation, proceed as follows:

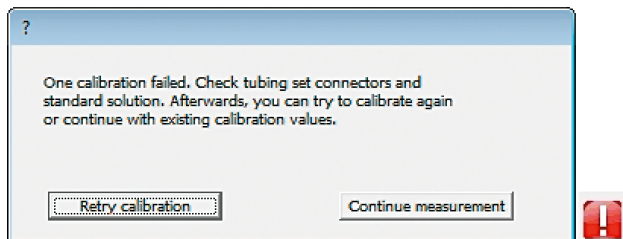
- ▶ Immediately stop the device!
- ▶ Secure the device against unauthorized operation.
- ▶ Check the malfunction against the “Table of error codes” from “10.2 Errors and Possible Causes” and “10.3 Error Message: “Calibration failed” ”.
- ▶ Only carry out the activities for troubleshooting described under the “10.2” and “10.3” or contact the Sartorius Service.
- ▶ If this does not solve the problem, contact the Sartorius Service.

10.1 Error messages

The device has some internal tests and gives appropriate error messages. The acknowledgment by the “OK” button closes these messages. As long as the error is not resolved, corresponding marks  and  appears on the control.

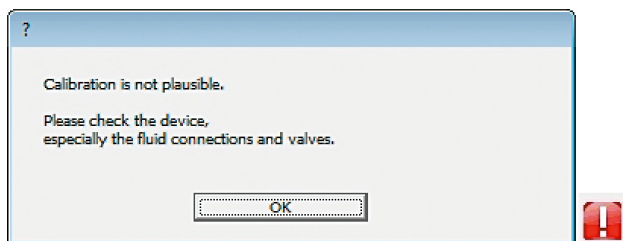
Pressing on these markers, the error messages can be opened again. If the error is resolved, the messages and marks will be removed.

10.1.1 Calibration Error



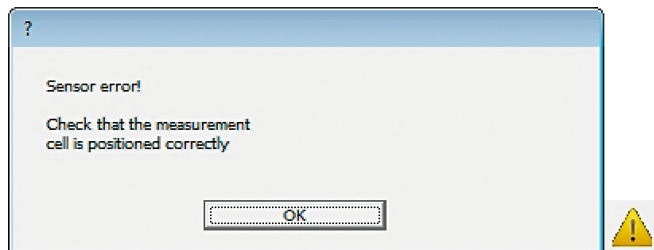
For each calibration, the resulting measurements must lie within given tolerances, otherwise this error message appears. Both tables (see "10.2 Error and Possible Causes", page 122 | "10.3 Error Message: "Calibration Failed"", page 124 give an overview of possible causes and repair.

10.1.2 Calibration Implausible



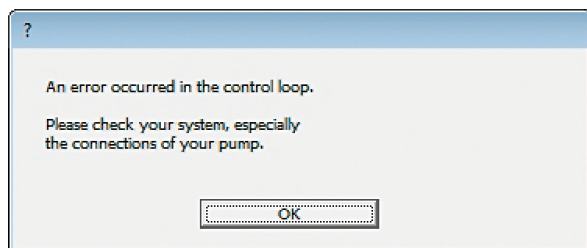
This test is a plausibility check, in which, although the calibration is running smoothly, false results occur (for instance if the standard concentrations are typed in wrong). For each calibration, the resulting values (offset | slope) must lie within given ranges, otherwise this error message appears. Both tables (see "10.2 Error and Possible Causes", page 122 | "10.3 Error Message: "Calibration Failed"", page 124 give an overview of possible causes and repair.

10.1.3 Sensor Error



The device is automatically monitoring if the pin connectors have electrical contact to the measurement cell. If this is not the case, erroneous readings may result. Please check whether the cell has been inserted correctly.

10.1.4 Error in Control



This message appears if the communication with an external digital pump is disturbed.

10.2 Error and Possible Causes

Error Error message	Possible cause
Calibration failed	<ul style="list-style-type: none"> – Calibration standard empty – Transport buffer empty – Air bubble inside the measuring chamber – Incorrect tubing set used – Conductivity cell not properly inserted – Wrong method selected – Valve tubes inserted incorrectly or not inserted – Dialysis only: three-way stopcock incorrectly adjusted – Tubing set out of date – Leaks in the tubing set – Calibration line blocked or tubing set clogged – Software versions of device PC not compatible – Valve (yellow) cannot be triggered
Measured value too high or too low	<ul style="list-style-type: none"> – Calibration erroneous – Parameter entry – “Standard” parameter entry and actual standard do not match – Air bubble inside the measuring chamber – Transport buffer empty – Leaks in the tubing set – Dialysis only: Reference measurement not performed – Dialysis only: Reference measurement not up-to-date

Error Error message	Possible cause
No or negative measured value	<ul style="list-style-type: none"> – Conductivity cell not properly inserted – Transport buffer empty – Tubing set clogged – Leaks in the tubing set – Tubing set out of date – No sampling probe or wrong sampling probe connected
Measured value varies greatly	<ul style="list-style-type: none"> – Air bubble inside the measuring chamber – Conductivity cell not properly inserted – Tubing set out of date
No connection to trace_mon	<ul style="list-style-type: none"> – IP address set incorrectly – PC: IP address not automatically assigned – Incorrect or defective power cord – Device boots but program does not start
Pump is not turning	<ul style="list-style-type: none"> – Remove pump cassette and clean motor shaft – Pull tight pump tube
No flow	<ul style="list-style-type: none"> – Transport buffer empty – Calibration standard empty – Tubing set clogged – Leaks in the tubing set

10.3 Error Message: “Calibration Failed”

Possible cause	What should you do?
1. The transport buffer is empty (Is there still enough transport buffer in the storage vessel? Is the suction tubing still in the storage vessel? Is buffer dripping into the drip counter?)	Connect new transport buffer and fill the tubing set.
2. A calibration solution is empty.	Connect new calibration solution.
3. Silicone tubes are not properly inserted into the valves.	Check if the valve tubes are seated properly and correct as required.
4. There is an air bubble inside the measuring chamber.	Restart the device and fill again.
5. A tube is pinched (e.g. when screwing the covers into the standard bottles.)	Remove pinch.
6. There is clogging in the tubing set (When the buffer remains in the tubing set over an extended period of time, salts begin to crystallize out. Crystallization usually occurs on the silicone tubes in the valves. Clogging can also occur due to microbial growth.)	Remove clogging. If this is not possible, use a new tubing set.
7. Leaks (If the clog is not removed, tubes may burst.)	Correct leaks. If this is not possible, use a new tubing set.
8. Sensor does not have proper contact (When the sensor is not inserted properly, the reverse side of the sensor does not have proper electrical contact with the sensor holder.)	Remove sensor and reinstall it properly.

	Possible cause	What should you do?
9.	The dialysis membrane was installed in the probe in the incorrect position (only for DIALYSIS application).	Correctly install a new dialysis membrane into the probe.
10.	Three-way stopcock was not properly adjusted (only for DIALYSIS application. An external sample was measured and the three-way stopcock was not reset).	Adjust the three-way stopcock to the correct position.
11.	The software versions of the BioPAT® Trace Multi Trace and the PC are not compatible.	Use suitable software. If necessary, reinstall the PC software "trace_mon" from the website.

11 Transport

The BioPAT® Trace | Multi Trace including accessories is delivered in protective packaging.

- ▶ Please save this packaging; proper (return) shipping is only possible in the original packaging.
- ▶ Upon receipt, check the delivery for completeness and any possible damage that may have occurred in transit.
- ▶ Any transport damage must be reported within a week of delivery. Complaints made after this date will not be accepted.

12 Storage and Shipping

12.1 Storage

- ▶ The device has been decommissioned.
- ▶ Clean the device.
- ▶ Store the device under the prescribed ambient conditions.

12.2 Returning the Device and Parts

Defective devices or parts can be returned to Sartorius. Returned devices must be clean, decontaminated, and properly packed.

Transport damage as well as measures for subsequent cleaning and disinfection of the device or parts by Sartorius shall be charged to sender.

Devices contaminated with hazardous materials, e.g. harmful biological or chemical substances, will not be accepted for repair or disposal. The devices must be decontaminated before shipping (see Chapter “13.1 Information on Decontamination”, page 128).

- ▶ Decommission the device.
- ▶ Contact Sartorius Service for instructions on how to return devices or parts (please refer to www.sartorius.com).
- ▶ Pack the device and its parts properly for return.

13 Disposal

13.1 Information on Decontamination

The device does not contain any hazardous materials that would necessitate special disposal measures. The cultures and media (e.g. acids, bases) used during the fermentation processes are potentially hazardous materials that could cause biological or chemical hazards.

According to the EU directives [European directive on hazardous substances], the owners of devices that come into contact with hazardous substances are responsible for properly disposing of these devices and must declare such devices when transporting them.

WARNING

Risk of injury due to contaminated devices!

Devices contaminated with hazardous materials (NBC contamination) will not be accepted for repair or disposal.

13.2 Decommissioning the Device

Procedure

- ▶ Switch off the device (see Chapter “7.2 Decommissioning”, page 74).
- ▶ Clean the device.
- ▶ Decontaminate the device.

13.3 Disposing of the Device and Parts

13.3.1 Information on Disposal

The device and its accessories do not belong in your regular household waste, since they are made of high-grade materials which can be recycled and reused. All parts must be disposed of properly by disposal facilities.

The packaging is made of environmentally friendly materials that can be used as secondary raw materials.

13.3.2 Disposal

Requirements

The device has been decontaminated.

Procedure

- ▶ Dispose of the device. Follow the disposal instructions on our website (www.sartorius.com).
- ▶ Dispose of the packaging in accordance with local government regulations

14 Technical Data

14.1 Measuring Data

	Value
Measuring principle	Enzymatic-amperometric
Measuring range (glucose)	0.10 – 40 g/L
Measuring range (lactate)	0.05 – 5 g/L
Measuring range (methanol)	0.5 – 20 g/L
Measuring range (ethanol)	1 – 40 g/L
Measuring frequency	up to 30 measured values/h
Measurement deviation (glucose)	≤3% at 5.0 g/L
Measurement deviation (lactate)	≤3% at 2.5 g/L
pH range of the medium	4.8 – 9.2
Service life of the enzyme electrode	30 days or 5000 analyses

14.2 Dimensions and Weights

	Value
Dimensions in mm (W × H × D)	120 × 170 × 200
Weight	1.8 kg

14.3 Installation Conditions

14.3.1 Ambient Conditions

	Value
Temperature	15 – 35 °C
Air humidity	10 – 90%

14.3.2 Footprint

	Value
Even surface	
Lateral distance	0.5 m

14.3.3 Storage Conditions

	Value
Temperature	15 – 35 °C
Air humidity	10 – 90%

14.4 Electrical Data

14.4.1 Power Supply

	Value
Analog output	0 - 10 V 0 - 20 mA 4 - 20 mA
Input voltage	100 - 120 220 - 240 V~ 50/60 Hz

14.4.2 Interfaces

	Value
Interfaces	RS 232, Ethernet, OPC UA, Modbus TCP
PC for controlling the operating system	Windows XP, Windows 7, Windows 8, Windows 10

15 Conformity Documents

The attached documents declare the conformity of the device with the designated directives or standards.



Original

EG-/EU-Konformitätserklärung
EC / EU Declaration of Conformity

Hersteller Sartorius Stedim Systems GmbH
 Manufacturer 34302 Guxhagen, Germany

erklärt in alleiniger Verantwortung, dass das Betriebsmittel
declares under sole responsibility that the equipment

Geräteart **Messgerät zur Überwachung | zur parallelen Überwachung von Bioprozessen**
 Device type ***Analyzer for monitoring | for parallel monitoring of bioprocesses***

Baureihe **BioPAT® Trace | BioPAT® Multi Trace**
 Type series

Modell **BPT0001 | BPT00M**
 Model

in der von uns in Verkehr gebrachten Ausführung allen einschlägigen Bestimmungen der folgenden Europäischen Richtlinien entspricht und die anwendbaren Anforderungen folgender harmonisierter Europäischer Normen einschließlich deren zum Zeitpunkt der Erklärung geltenden Änderungen erfüllt:

in the form as delivered fulfils all the relevant provisions of the following European Directives and meets the applicable requirements of the harmonized European Standards including any amendments valid at the time this declaration was signed listed below:

2014/30/EU Elektromagnetische Verträglichkeit / *Electromagnetic compatibility*
 EN 61326-1:2013

2011/65/EU Beschränkung der Verwendung bestimmter gefährlicher Stoffe in Elektro- und Elektronikgeräten (RoHS) / *Restriction of the use of certain hazardous substances in electrical and electronic equipment (RoHS) in conjunction with 2015/863/EU and 2017/2102/EU*
 EN 50581:2012

Die Person, die bevollmächtigt ist, die technischen Unterlagen zusammenzustellen:
The person authorised to compile the technical file:

Sartorius Stedim Systems GmbH
 Product Compliance
 34302 Goettingen, Germany

Sartorius Stedim Systems GmbH
 Guxhagen, 2020-07-30

Dr. Thorsten Peuker
 Managing Director

Volker Otto
 Head of BPS Operations, Guxhagen



Original



UK Declaration of Conformity

Manufacturer Sartorius Stedim Systems GmbH
Robert-Bosch-Str. 5-7
34302 Guxhagen, Germany

declares under sole responsibility that the equipment

Device type Analyzer for monitoring | for parallel monitoring of bioprocesses

Type series **BioPAT® Trace | BioPAT® Multi Trace**

Model **BPT0001 | BPT00M**


in the form as delivered fulfils all the relevant provisions of the following British Regulations and meets the applicable requirements of the British Designated Standards including any amendments valid at the time this declaration was signed listed below:

	EMC	RoHS
Regulation	The Electromagnetic Compatibility Regulations 2016 UK Statutory Instruments 2016 No. 1091	The Restriction of the Use of Certain Hazardous Substances in Electrical and Electronic Equipment Regulations 2012 UK Statutory Instruments 2012 No. 3032
Standard(s)	BS EN 61326-1:2013	BS EN IEC 63000:2018

The person authorised to compile the technical file:

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Sartorius Stedim Systems GmbH
Guxhagen, 2022-10-04

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Manager - Product Compliance BPS Systems
Operations – BPS Systems

16 Licensing or Trademark Information

GNU Licensing

The software code for the DCU systems is partially subject to the license terms of the “GNU General Public License (GPL)” or the “GNU LESSER General Public License (LGPL)”.

If applicable, the license terms of the GPL and LGPL as well as information about the options for access to GPL code and LGPL code used in this product are available upon request.

The GPL code and LGPL code contained in this product are published without any guarantee and subject to the copyright of one or more authors.

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Sartorius reserves the right to make changes to the technology, features, specifications and design of the equipment without notice.

Masculine or feminine forms are used to facilitate legibility in these instructions and always simultaneously denote all genders.

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Last updated:

11 | 2022

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