<u>User's guide – installation of pH-stat module</u>

The module is intended for pH-stat cultivation which is driven by automatically controlled CO₂ sparging. The module includes the Peristaltic Pump PP600 and an assembly of the Gas valve and Flow control valve (pH-stat kit). Supporting control software and additional tubing are needed (Parker polyflex presto TPU 6x1 tubing is delivered with the pH-stat module and silicone tubing Ø 6/3 mm is delivered in the spare kit with the Photobioreactor FMT 150). The CO₂ and Air sources for sparging of the biomass are not included.

Use the Peristaltic Pump **PP600** and an assembly of the Gas valve and Flow control valve (pH-stat kit) that are shown in the pictures below.

The Gas valve together with PP600 is intended for CO_2 flow regulation (ON/OFF) depending on the target and actual pH value of the suspension. Once a higher pH than the set value is detected then the PP600 opens the Gas valve for CO_2 sparging. The Flow control valve allows you to lower or increase the added CO_2 flow manually. This allows controlling the level of pH drop after the CO_2 addition to the suspension. The Flow control valve has to be open partially at the beginning of the experiment.





Installation and control

- 1. Connect the power supply cord of PP600 to the "POWER" port.
- Connect the PP600 with the PBR: plug one end of the communication cable (AUX cable) into the "AUX IN" port of the PP600 and the other end into the "AUX 1" port on the rear side of the Photobioreactor.
- 3. Turn on the Main switch.

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4. Connect the pH-stat kit with the PP600: plug the cable into the "VALVE 1" port on the rear side of the PP600.



5. Connect the Parker tubing for CO₂ and Air delivery as shown in the picture below. Use the input gas pressure around 1 bar.

In case a pressurized source of CO_2 or Air will be used it is necessary to lower the outcoming gas pressure via any pressure-reducing valve to the appropriate level before connecting the gas source to the pH-stat kit.



6. Connect the output Parker tubing containing the final mixture of CO₂ and Air via the silicon tubing with the bubble interruption valve and humidifier as shown in the picture below.



1) Bioreactor.

2) Source of CO_2 for a pH-stat regulation by CO_2 (any CO_2 addition to the biomass suspension results in pH drop). CO_2 is added automatically in dependence on the actual measured pH of biomass suspension in the cultivation vessels.

3) Pressure reducing valve or regulator.

Pump (4) controls the CO_2 addition automatically by opening and closing the gas valve (5) if the CO_2 addition is needed. Valve (6) is manual and controls the CO_2 flow rate.

7) Source of Air or any other bubbling gas (for example air pump or any air compressor or any other gas from pressure cylinder...). It is always necessary to reduce the outcoming pressure from the pressure cylinder to approximately 1 bar or less. The gas from this source is intended for the continuous bubbling of the biomass. The bubble interruption valve **8)** stops the bubbling only for OD measurement.

9) Filter.

10) Humidifier.

11) Waste bottle.

WARNING: Don't connect CO₂ and Air tubing in other way! A wrong connection can cause a damage because of over-pressurizing or flooding the system.

- <u>The setting of PP600</u>: It is necessary to set the corresponding ID for the used PP600. For this
 purpose, use the control display of PP600 and go to Menu >> Setting >> Pump-ID. Choose [1] that is
 intended for pH regulation via acid.
- The setting of the control PBR's software: Select pH Regulator acid in the Protocol folder. Choose: Mode = Gas valve and set the target pH value. Upload the setting by "Upload" button.

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In the case of running the turbidostatic cultivation without pH-stat, there is no need for 2), 3), 4), 5), or 6) at all. Only the connection of a bubbling gas (for example 5000 ppm CO₂) through the bubbling interruption valve 7) is needed.