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Master Thesis in

European Master in Nuclear Applications

Setup of a gas exchange system for the application of radioactively labeled CO_2 on plants

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12 August 2013

I hereby declare that I have written the Master Thesis on my own and have used no other than the stated sources and aids.

Signature………………………………………………………………….

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1. Introduction

Nowadays agriculture faces major challenges. It is assumed that the need for plant material in form of food, feed and raw material will have doubled until 2050 (FAO, 2009). This is mainly due to the growing world population. At the same time, the growing of plants becomes increasingly difficult in some parts of the world due to environmental changes. These include drought stress, water flooding, soil nutrient depletion, erosion and pollution (FAO, 2009). This requires a more efficient and sustainable plant production with less resource demand. The problem is more complex than it appears in the first place. There is no plant species, which shows optimal performance in any environment and, thus, specific plant traits have to be identified that are individually adapted for the different regions of the world. The key to it is phenotyping. Phenotyping is the study of plant structures, functions and responses to environmental conditions based on its genome. All measured parameters more or less come down to the same question: How much biomass is accumulated by a plant (harvesting yield) and how efficient does the plant use the available resources?

One important function with respect to harvesting yield implies how quickly and efficient the plant takes up carbon dioxide (CO_2). CO_2 is the main source for any synthesis of organic molecules in plants. It is taken up through highly controlled pores in the leaf called stomata. With the help of enzymes CO_2 is fixed and converted to carbohydrates, mostly sucrose, via photosynthesis (see Figure 1). Inside of the leaf, the photoassimilates (the products of photosynthesis) are either stored as starch or loaded into the phloem for further long distance transport to so-called sink organs (roots, shoots, etc.). All the involved metabolic processes like photosynthesis, respiration, loading, transport and unloading of phloem very much depend on several conditions and need to be quantified for each phenotype.

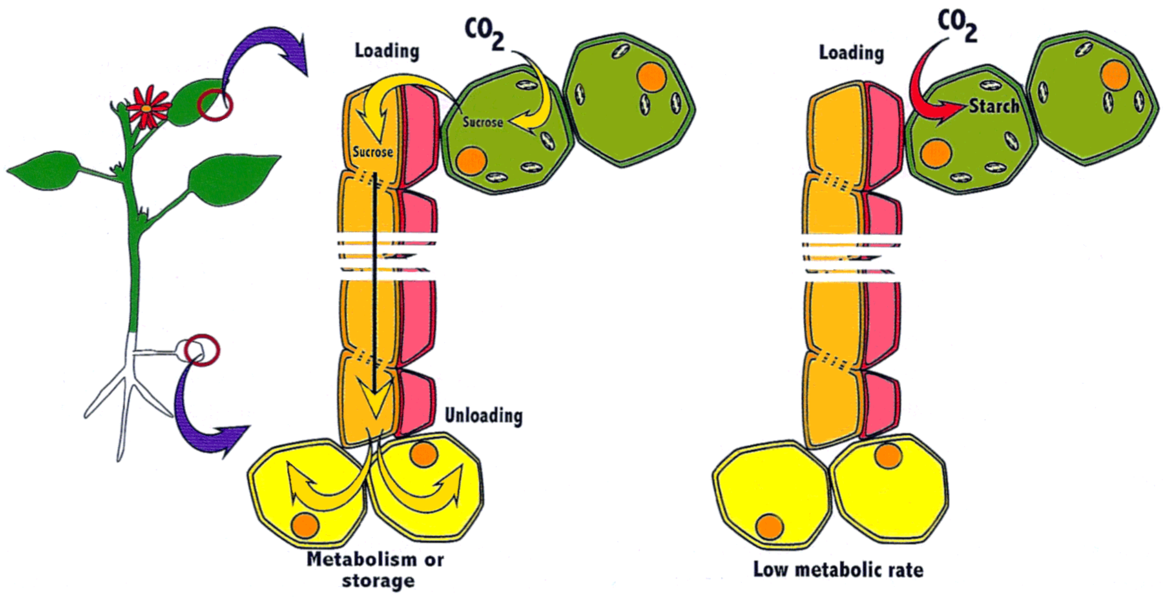


Figure 1: Assimilation of CO_2 in plant leaf, translocation to sink tissues & allocation of sucrose (Scott, 2008)

Whereas the analysis of structural propertied of a plant is relatively straightforward, functional parameters of plants require more sophisticated and often non-invasive methods. The Institute of the Bio- and Geosphere, IBG-2: Plant Sciences, at Forschungszentrum Jülich, Germany, employs tracer technologies to study CO_2 uptake, transport and storage of products of photosynthesis dynamically in vivo. For this purpose, the CO_2 is labeled with radioactive ^{11}C (a positron-emitter with a half-life of 20 min) and administered to the plant through the leaf. In order to administer a defined gas composition (containing also the radioactive CO_2) and monitor the gas exchange with the plant leaf, a special gas exchange system for atmospheric conditions is required. The scope of this master project is to partially setup a gas exchange system, which can deliver different gas compositions and is also dedicated for radiotracer applications, and evaluate its functionality. However, a test with a plant is not performed, since this will go beyond the scope of this thesis.

2. Functional principle of the gas exchange system

A plant leaf is on constant exchange with its gaseous environment, where the exchange of a specific gas is achieved by diffusion. In simplified terms, a plant takes up gases (like CO_2 and O_2) and releases or fixes them depending on the concentration gradient between leaf and environment. During daylight, the plant is net taking up CO_2 and releasing O_2 and water vapor (H_2O) due to photosynthesis (Figure 2). These gas exchanges take place under ambient pressure. Any change in pressure directly influences the diffusion gradients and hence the uptake of the gases.

Figure 2: simplified gas exchange of a plant leaf during daylight

Basically, the task of the gas exchange system is to provide a defined gas composition (air with different concentrations of CO_2) at atmospheric pressure and to monitor the CO_2 uptake and the water transpiration, simultaneously. The design of the gas exchange system is based on a previous system described in (Jahnke, 2001) with slight modifications. The following list gives an overview about basic requirements to be met:

- The system should be applicable as open and closed system. In the open system a fresh gas composition is delivered to the plant, the exchange with the plant is measured and the gas is leaving the system at some point. In the closed system, no fresh gas will be delivered to the plant, which means that the plant is exposed to changes in gas composition due to photosynthesis, photorespiration, and dark respiration by the enclosed leaf.
- In order to imitate natural gas exchange conditions (as with the other leaves of the plant), the pressure inside of the system needs to be controlled.
- The gas delivered to the plant is composed of air with specific concentrations of CO_2, N_2, O_2 and water vapor. These concentrations should be variable to study the response of the plant to environmental changes (e.g. elevated CO_2 concentration).
- The CO_2 concentration and the water content are measured with an accuracy of $\pm$ 1 ppm. For differential measurements it should ideally be in the 1/10-ppm range.
- There is demand for several by-pass systems for flow matching.
- For safety reasons, no radioactive gas should leave the system, not even through a mistake or failure. This also entails that all parts should be as leak tight as possible, that CO_2 inert, clean and smooth materials should be used, and that dead volumes should be avoided. This minimizes adsorption of the CO_2 and long retention times of the radioactive material.
- The system should be modular to allow changes afterwards.

However, there are some constraints for the setup. Available equipment is to be used as far as possible. And all active components are to be controlled remotely by a LabVIEW program (National Instruments). The following sections will discuss the functional design in detail.

2.1. The Main Gas Cycle

Figure 3 presents an overview of gas flow in the system. Starting from the left bottom, a defined gas composition is delivered by the Gas Supply (blue box) and the Gas Conditioning & Mixing modules (turquoise boxes). The Gas Conditioning & Mixing module is discussed in section 2.2 because it needs detailed consideration. In order to switch quickly between different kinds of gas composition, at least two of such modules are to be implemented into the system. The selection of either gas composition is controlled by multi-valve MV 4.1. The Growth Chamber is not part of the actual gas exchange system, but would enable the sampling of air from the place where the plants are grown. After passing a filter (FI 4.1), the gas stream is divided into a main flow way and a branching way. The gas of the main flow way is lead to the plant leaf in the Leaf Chamber (section 2.3) by passing the 4/2 way ball valves MV 5.1 and 5.2. Pump GP 4.1 controls the mass flow, which is measured by the mass flow meter MFM 5.1. The gas pump GP 8.1 helps to maintain atmospheric pressure in the Leaf Chamber, which is monitored by a pressure sensor in the Leaf Chamber (Pd 5.1 - not shown). The transpiration of the plant can be monitored by the humidity sensors H 5.1 and H 5.2 (inter alia). A dew point trap DP 8.1 can optionally remove the humidity transpired by the plant in the Leaf Chamber by turning the 3/2 way multi-valves MV 8.1 and 8.2. The branching part from the initial reference gas stream (delivered by pump GP 4.2) is now compared to a fraction of the gas stream from the Leaf Chamber by a differential IR gas analyzer (IRGA-diff). It measures the CO_2 concentration as well as the absolute humidity in two cuvettes (reference, Ref, and measure, Meas). It also delivers an input reference value for the previously mentioned dew point trap DP 8.1. On demand, the differential IRGA cuvettes can be matched by switching the 3/2 way multi-valve MV 8.3. It should be ensured that the pressure measured by differential pressure sensor Pd 8.1 does not exceed a certain level, since the IRGA is pressure sensitive. The gases are then directed to the Gas Exit and Absorption, which is treated in section 2.4.

In order to allow a closed cycle, the multi-valve MV 5.1 is switched by which the gas supply and measuring devices in the main cycle are decoupled from the Leaf Chamber. The feedback loops for GP 8.1 and DP 8.1 should be interrupted, and the devices should run with a default value. By switching multi-valve MV 5.2, the gas from the Leaf Chamber also passes the Radioactive Tracer Application Module. It comprises its own measurement devices and actuators (pumps and administration kits). But it is not part of this work (for further information see attachment).

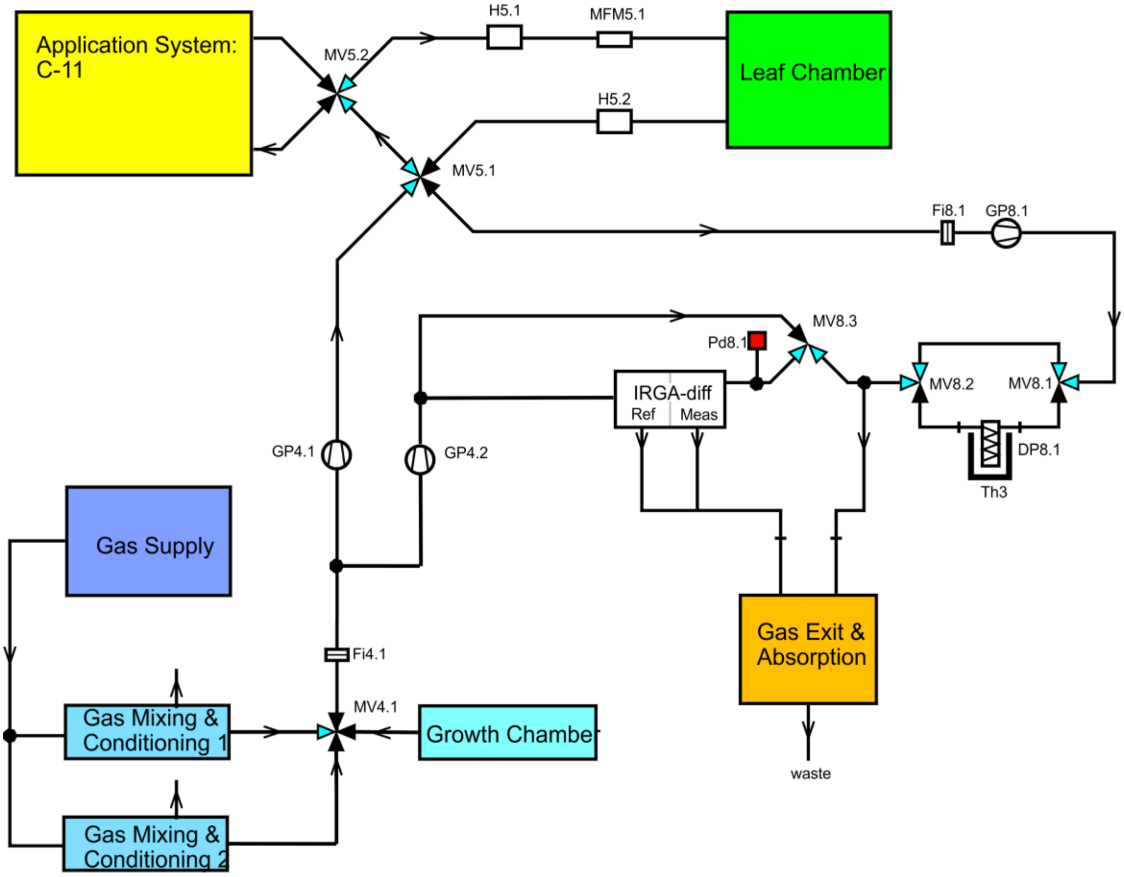


Figure 3: Gas flow chart for the Main Gas Cycle

2.2. The Gas Mixing and Conditioning

The modules Gas Mixing & Conditioning 1 and 2 are identical in function. Figure 4 serves as detailed flow chart of both modules, but exhibits the naming for module 1. In module 2, all component numbers will start with a 2.

Four gas lines provide clean and dry gases, three of which are produced synthetically (N_2, O_2, CO_2). The fourth gas line delivers CO_2 free and dry air, produced by a Gas Generator, which adsorbs CO_2 in compressed air. A cylindrical tank homogenizes the gas before it is processed. The gas is delivered in two modes: either the CO_2 free air is combined with CO_2 or a mixture of N_2 and O_2 is combined with CO_2. The mode of gas mixture is controlled by the valves V 1.1 to V 1.4. Because the CO_2-free air together with the CO_2 will be used most of the time, the valves V 1.2 and V 1.3 are of the type normally open. The valves V1.1 and V1.4 are normally closed. Mass flow controllers (MFC 1.1 – 1.3) set the exact amount of each gas. The gases are mixed in a way, that the high gas stream (air or N_2) does not hamper the low gas flow (CO_2) of entering the main flow tube. Since the air is completely dry, it has to be moistened by a humidifier and set to a defined dew point. The humidity is not subject to a control loop here, so the dew point system should work stable. For some purposes, it might be useful to by-pass the humidifying unit, which is done by the 3/2 way multi-valves MV 1.1 and 1.2. A CO_2-absorber can be included optionally. A small spillover is required to release gas, which is not needed. Therefore, the differential pressure in the system should be slightly positive towards atmospheric pressure. Otherwise it might happen that air is sucked in at the spill-over. To ensure a correct operation the differential pressure is monitored by pressure sensor Pd 1.1 which gives an alarm if the differential pressure is out of range.

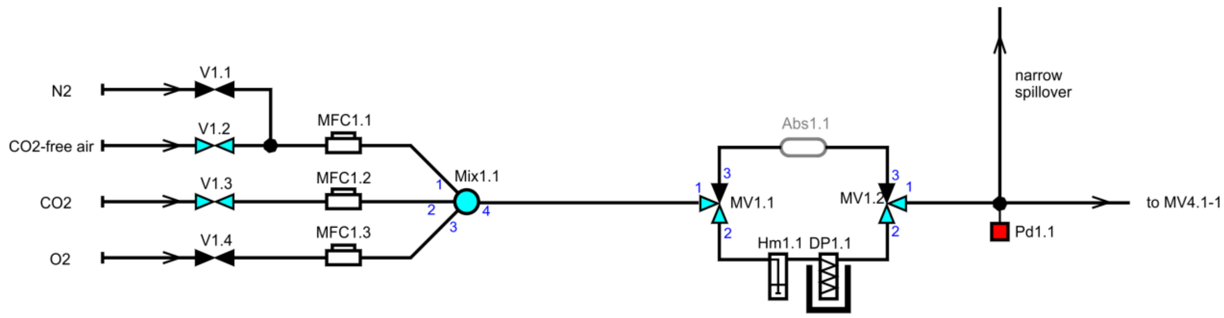


Figure 4: Gas flow chart of gas mixing and conditioning unit

2.3. The Leaf Chamber

The Leaf Chamber is a flat container, into which an attached leaf is inserted (similar to Figure 5). It consists of a frame and transparent glass lids to allow illumination. The petiole is passed through an opening and sealed with putty. The Leaf Chamber has an inlet and an outlet for the gas, and a capillary connection to the pressure sensor Pd 5.1, which was already mentioned in section 2.1. The differential pressure inside the Leaf Chamber is kept zero by the aid of pump GP 8.1 of the main gas cycle as it was explained before. However, the Leaf Chamber is considered to be the leakiest part inside the system. Keeping the differential pressure zero also reduces the risk of releasing radioactive $^{11}CO_2$ or sucking in gas from outside.

The Leaf Chamber can be disconnected from the remaining system by closing valves V 5.3a and V 5.3b (normally open). For direct radioactive $^{11}CO_2$ application, it should be possible to attach a CO_2 trap through quick connectors. A small pump (GP 5.2) will transport the radioactive gas to the leaf. Valves V 5.4a and 5.4b close the inlet of the quick connectors, if no CO_2 trap is attached. The valves are normally closed. Eventually, a ventilation module, which is supposed to apply some “wind” to the plant leaf, will expand the Leaf Chamber unit. But this will not be part of this work as everything, which is grayed out.

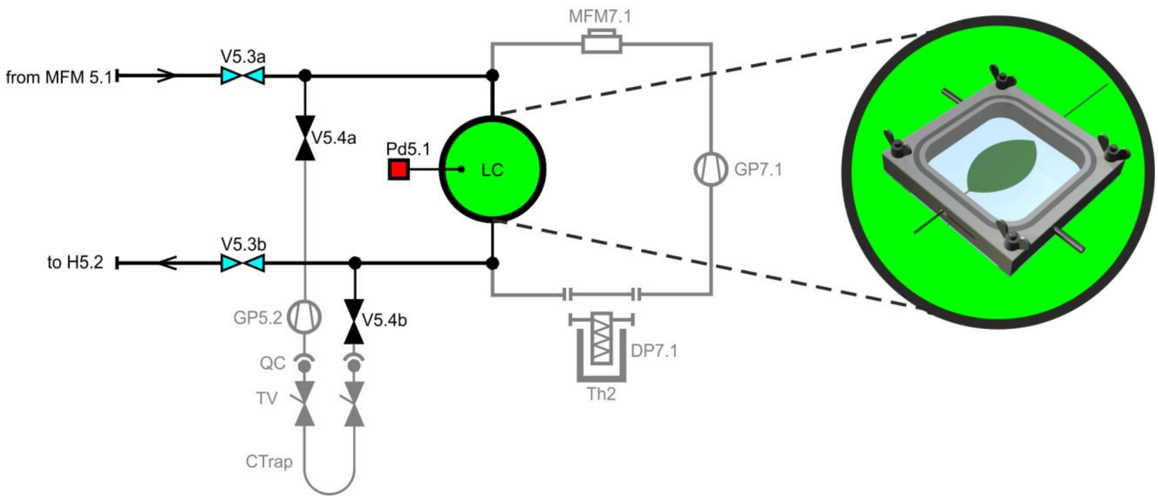


Figure 5: Gas flow chart of the Leaf Chamber unit

2.4. The Gas Absorption and Exit

The gas of the Main Cycle is entering the Absorption and Exit unit from two sides, the IRGA and MV 8.1. Both are merged in a mixing point PRP, where air from the surroundings is drawn in. In order to ensure that no radioactive gas leaves through the air inlet, a pressure sensor Pd 9.1 monitors the differential pressure and a gas pump GP 9.1 is regulated accordingly. For security reasons, the air, which is sucked in, passes a CO_2 absorber Abs 9.1 in case of a failure. Downstream the mixing point, radioactive $^{11}CO_2$ can be fixed on CO_2 absorbers and measured for radioactivity or allowed to decay. The inflow is governed by valves V 9.2a and 9.2b, or 9.3a and 9.3b. If no tracer is administered, the gas can exit the system by opening valve V 9.1 and a manual valve (NV 9.1). All valves in this unit are normally closed, so that in case of a power failure, no radioactive gas can leave the system. The Gas Pump GP 9.1 should not run, if all valves are closed.

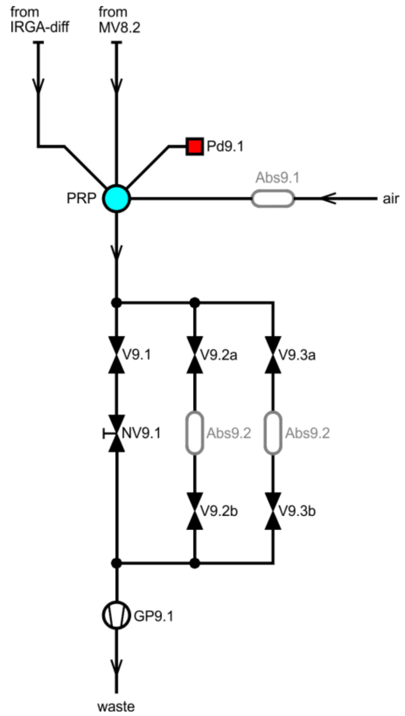


Figure 6: Gas flow chart of the Gas Absorption & Exit Unit

3. Summary of preliminary work

Prior to the master project, some work was performed as part of a mini-project. In a first step, technical equipment had to be assigned to the functions stated in the flow charts. In doing so, missing equipment had to be identified and procured. In a second step, PC connections with miscellaneous devices were established and LabVIEW programs were written dedicated to test their performance and suitability. As follows from the previous chapter, three basic physical values need to be controlled: the mass flow inside of the main cycle, the pressure at several positions and the humidity on demand. Each of these control loops had been tested individually in simple setups. This list gives an overview about the tests that have been done:

- An available membrane pump type (NMP850, KNF Neuberger, Germany) was tested for loss of CO_2 (leakage test) and found to be suitable.
- A small control loop for regulating the mass flow and the differential pressure with two pumps was set up and its performance was evaluated.
- A suitable humidity sensor was found and a casing for it was constructed
- A simple setup for proper humidification of the air was tested.
- Performances of two potential systems as dew point traps were measured and assigned to a suitable location within the system according to their dynamic properties

The following sections explain the tests in more detail. The leakage test will not be explained any further since it proved the general applicability of the membrane pumps (leak tightness suitable).

3.1. Pressure and mass flow rate regulation

As was mentioned in the previous chapter, a crucial part in setting up the gas exchange system is the stability of the flow rate and the pressure. A simplified gas flow chart for the control of flow rate and pressure is demonstrated in Figure 7. On the input side, a gas pump GP 1 (representing gas pump GP 4.1) provides a gas flow measured by a mass flow meter MFM (M82088570, 5l/min Bronkhorst-Mättig, Germany), representing MFM 5.1. An increasing flow rate also results in an increase in pressure, which is monitored by a sensor PD (representing Pd 5.1). In order to provide atmospheric conditions, the pressure has to be controlled by a second pump GP 2 (representing GP 8.1). It is aiming for zero differential pressure by drawing air on the output side. In the course of this, the differential pressure cannot directly be measured in the gas line, since the membrane pumps lead to a pulsating gas flow and pressure. This results in large fluctuations in the differential pressure, interfering with the actual offset signal caused by the increased dynamic pressure. Connecting the differential pressure sensor PD to a 30 cm long capillary with an inner diameter of 0.2 mm solves this problem (see Figure 8). The sensors in use have a range of ± 70 mbar (CTEM7N070GY0, Sensortechnics, Germany) and require a connection adapter made of stainless steel, which was already in place.

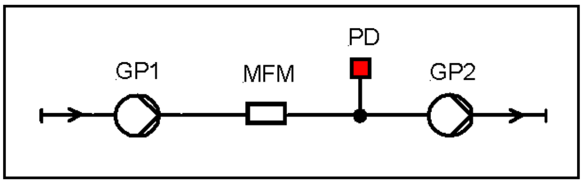


Figure 7: block diagram for testing the control loops for flow rate and pressure

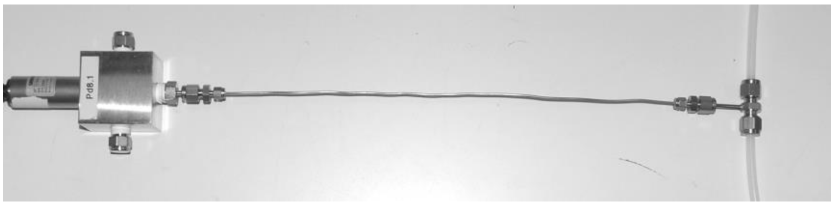


Figure 8: setup for measuring the pressure

Another issue is that both pumps influence each other, since a change in mass flow rate also results in a pressure change. The second pump reacts accordingly, increasing the mass flow rate, which slows down the first pump. Simply put, there's positive coupling of the two processes on each other, which means that one value represents a disturbance to the other process, respectively (see Figure 9). So, the control values had to be adjusted in such a way that there is no oscillation between either pumps or the second pump takes over completely.

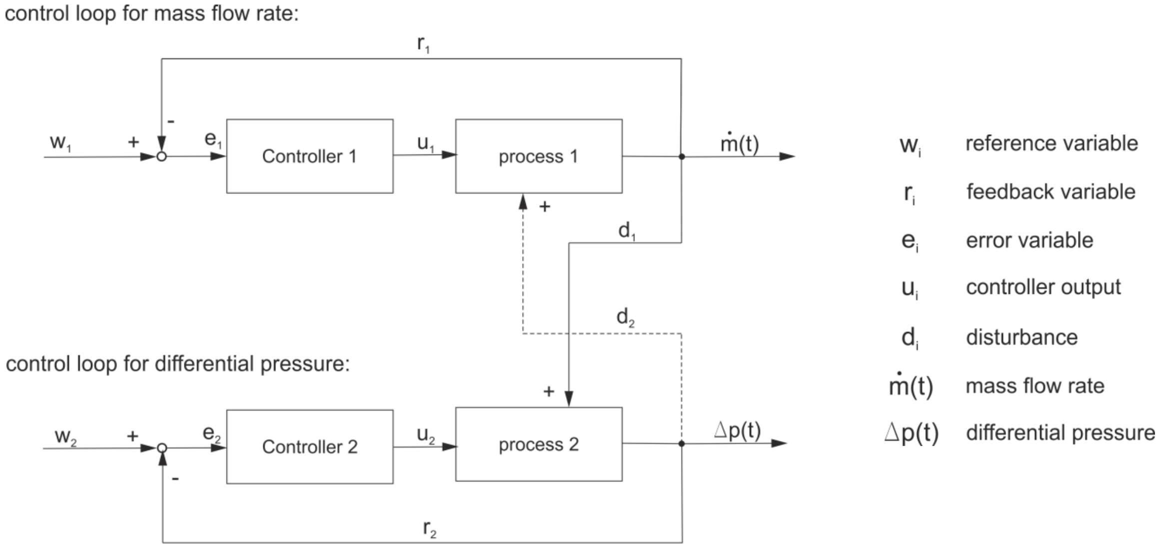


Figure 9: scheme for control loops for mass flow rate and pressure at Pd 5.1 and their influence on each other

Since the reference input (the mass flow rate) is assumed to be constant during a measurement and regulation of disturbances is the main issue, a common PID type controller was chosen. It is already implemented in LabVIEW and can quickly be installed in a program. Here, the derivative part was neglected because they are known to be sensitive to noise. However, the integral time is necessary to avoid a permanent offset. The control parameters for proportional gain K_p and integral time T_N were determined empirically based on guesses and are compiled in Table 1. These values are just valid for an intermediate region of flow rate and pressure. As both gas pumps affect each other, the second gas pump GP 2 reacts with a slight delay to the pressure change to avoid interaction. The values were not evaluated with an additional external disturbance and just serve as orientation. Moreover, it is assumed that they have to be adjusted when the complete system is setup and the flow resistance is higher.

	control loop for MFM	control loop for pressure
range	500 to 3000 ml/min	± 25 mbar
constant gain K_p	0.002	-0.003
Integral time T_N [min]	0.02	0.03

Table 1: preliminary control loop parameters for regulating mass flow rate and pressure

3.2. Humidification of the gas

The gas composition provided by the Gas Supply is completely dry, so it requires humidification. It is common to bring the humidity to 100 % saturation (Hm 1.1 and 2.1) and to use a dew point trap (DP 1.1 and 2.1) to adjust to a specific relative humidity. In order to prove the proper function of a humidification unit, first, a humidity sensor had to be procured. It will later be implemented into the system as H 5.1 and 5.2 to measure the transpiration of the plant with high accuracy. For this reason, a HygroClip HC2-IC102 (Rotronic, Germany) was chosen, which delivers accuracy of about ± 0.8 %rH (/± 0.1 K, response time τ_{63} without filter < 10 s). The sensors have the great advantage that they are applicable without transmitter and can just be connected to the computer via converter cable (AC3001, Rotronic, Germany) to a USB2 port. Additionally, LabVIEW programs (including its drivers) are freely distributed by the manufacturer and can be modified or implemented into other LabVIEW programs easily.

A slight drawback of the sensor is its size compared with the standard inner diameter of the tube. A cavity of about 15 mm diameter and 40 mm length is required to contain the sensitive part of the clip. For this reason a special casing made of stainless steel was designed (technical drawing see attachment) and machined. The sensor is brought into the casing via a Cable Gland (AC1301-M, M25x1.5, Rotronic, Germany), so that the sensor can be easily removed without the need of additional sealing (see Figure 10).

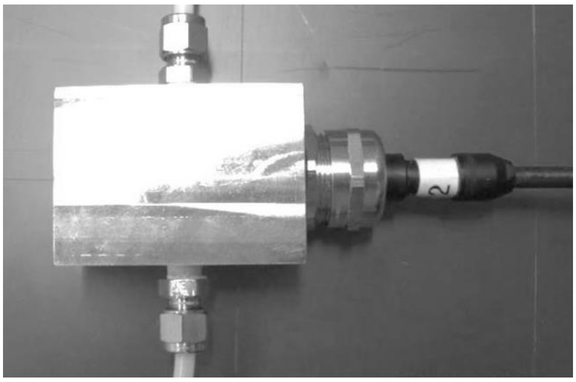
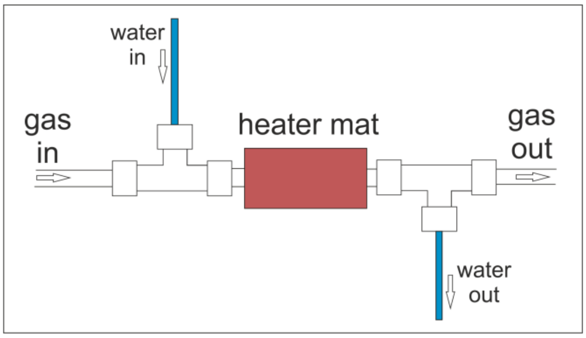


Figure 10: Humidity Sensor with Casing and Cable Gland

The humidification of the gas stream was realized in a simple setup: water is dripped into the gas line and heated in a short segment. The heating is provided by a flat heater mat of 15 cm x 5 cm size (Etched Foil Silicone Heaters, 245-556, RS, Germany), which is rolled around a stainless steel tube from the short end side and fixed with cable ties (Figure 11). The water inlet can be seen on the upper left. Remaining water leaves the tube at the lower right side in the test setup. Later the water outlet was combined with the dew point trap in order to remove the condensed water.



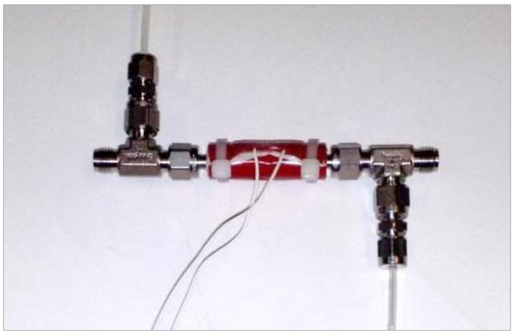


Figure 11: humidification unit: heater mat with water inlet and outlet, left: flow way, right: real setup

A peristaltic pump (ISM852, Ismatec, Germany) is used to transport the water from a reservoir to the gas line and back to the reservoir in a closed loop. The hose pump is not subject to remote control and the number of revolutions is kept constant. In this way, the humidity can be set to almost 100 %rH.

3.3. Choice of the dew point traps

In total four dew point traps will be needed for the gas exchange system: DP 1.1, DP 2.1 (gas mixing), DP 8.1 (main cycle) and later also DP 6.1 (as part of the radiotracer application module). These dew point traps can be implemented in two different ways. For each system two assemblies are available. The first dew point trap version (Figure 12 left side) consists of a cooling unit with a Peltier-element (DA-075-12-02, Laird Technologies, Sweden). The cooling unit is a hollow metal block directly attached to the Peltier-element. Inside, the gas stream flows along cooling ribs aligned in a U-shape. Condensing water is guided towards the bottom of the metal block and collected in a water reservoir. The collected water can be reused for humidification of the gas, building a closed loop. The hose pump is utilized again to transport the water back to the gas stream in front of the heater mat.

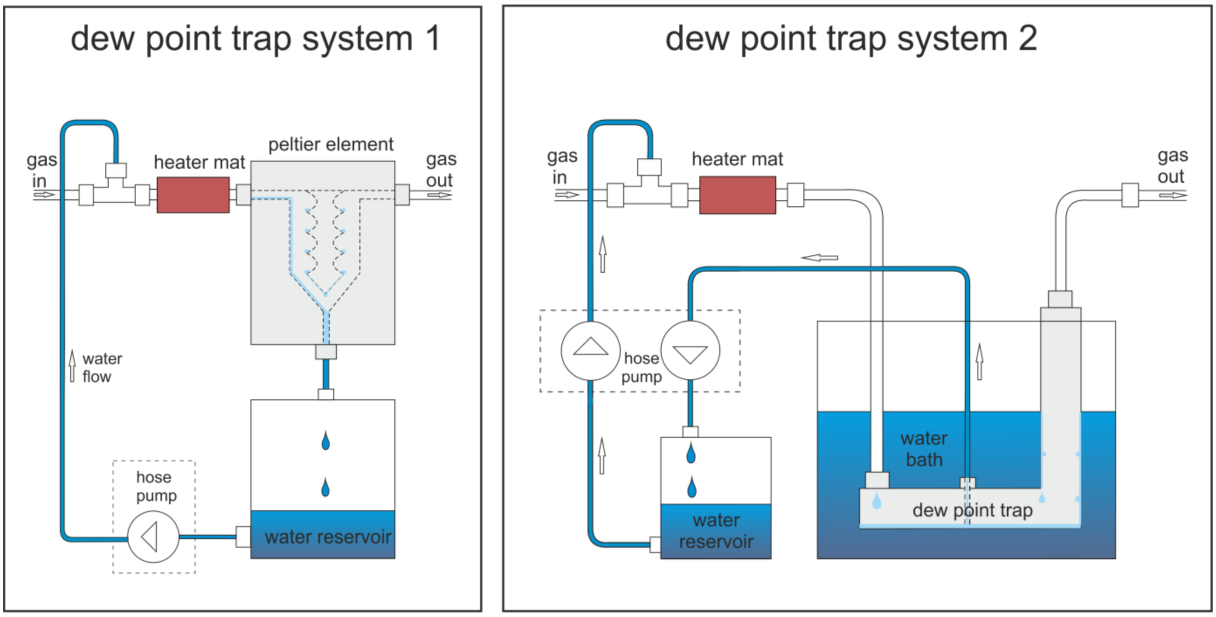


Figure 12: dew point traps: 1: Peltier-element and cooling unit; 2: custom-built dew point trap in water bath

In dew point trap version 2, a custom-built dew point trap (already available) is placed into a cooled water bath (refrigerating and heating circulator F32-HE, Julabo, Germany) as indicated in Figure 12 on the right. Basically, the dew point trap is an L-shaped metal tube with 2 cm inner diameter with connections for gas in and out at each end. Thereby, it is important that the upper part of the "L" is connected with the gas out tubing outside of the water bath, so that water drops, which might be built at the output side, are properly collected inside of the dew point trap. With the help of the hose pump and tubing, which reaches to the bottom of the trap, the condensed water is soaked up and funneled to a water reservoir. Again, this water can be brought back to the gas stream in a second loop with the same hose pump.

Both systems can be controlled remotely. Before, the Peltier-element was regulated by a controllable power supply (EA-PS 3032-10B, Germany), where the voltage is fixed to 12 V and the current is varied from 0 up to 7 A with the help of LabVIEW. But since two of these power supplies occupy a lot of space, smaller ones will replace them and temperature controllers will be introduced additionally.

Step responses were obtained to identify which system adjusts faster to changes and therefore is more suitable as DP 8.1 (and DP 6.1). For both systems, the reference relative humidity was set from 60 % to 40 % (cooling) and vice versa (warming). Since the dew point temperature ranges were comparable, they did not influence the choice of the dew point trap assignment.

First of all, it became obvious that both systems react differently if the reference humidity is increased by warming or decreased by cooling. By drawing a tangent in the inflection point, the equivalent dead time, T_u, and the settling time, T_g, were obtained for both systems for the warming phase as well as for the cooling phase. The results are listed in Table 2 (warming phase) and Table 3 (cooling phase). As the equivalent dead time and settling time of the Peltier-element are both smaller, the Peltier-element is found to be more dynamic than the refrigerating circulator.

warming:

	Peltier-element	Refrigerating circulator
K_P*	-13	40
T_u	0.3 min	0.6 min
T_g	3.1 min	19 min

Table 2: characteristics of Peltier-element and Refrigerating circulator for warming phase

cooling:

	Peltier-element	Refrigerating circulator
K_P*	-21	6
T_u	0.6 min	1.5 min
T_g	10 min	6.9 min

Table 3: characteristics of Peltier-element and Refrigerating circulator for cooling phase

Consequently, the Peltier-element will be used in the Main Gas Cycle (and Application module). Two sets of controller parameters are currently required for the Peltier-element, to compensate for the missing active heating and thus different dynamic behavior for the cooling and warming phase. The controller parameters for a PI controller were chosen according to Ziegler-Nichols (Lutz & Wendt, 2010) and are compiled in Table 4. They resulted in slight overshooting (not shown), which indicates that the parameters require optimization. As it was already mentioned for the control loops of mass flow rate and differential pressure, the found controller parameters need to be evaluated, once they are introduced into the complete gas exchange system. It is recommended to optimize the parameters according to Chien, Hrones and Reswick (Lutz & Wendt, 2010), in order to avoid overshooting of the Peltier-element. Furthermore the humidification segment has to be removed.

parameters	warming phase	cooling phase
K_P	-12	-20
T_N	1.2 min	1.7 min

Table 4: chosen parameters for a PI controller according to Ziegler-Nichols

The refrigerating circulator will be operated without feedback control of the humidity. It has a very stable behavior since the temperature of the water bath is controlled by the refrigerating circulator itself within the range of $\pm$ 0.01°C according to the specifications of the manufacturer. It will be used in the Gas Mixing and Conditioning modules 1 and 2.

4. Mechanical implementations

The gas exchange system is designed for 6 mm gas lines (= 4 mm inner tubing diameter). In order to have inert and flexible material, PTFE (Teflon) tubing is used. For gas lines with lower flow rates, such as branching lines, also PE tubing with a diameter of 3 mm is taken. The gas tubing is connected to the devices with the help of miscellaneous Swagelok fittings (Swagelok, Germany). If fittings have an NPT thread, the thread is additionally sealed with Teflon tape.

All devices, including data acquisition and processing (computer) are supposed to fit into a 19-inch rack with 200 cm height and 80 cm depth. Some devices already occupy a lot of space, so that the remaining space is very limited. Consequently, the arrangement of all other devices should be carefully planned. This chapter deals with the implementation of all devices and explains the difficulties. A complete device list can be found in the attachments. For now, the technical equipment, which is directly part of the gas flow charts, is summarized accordingly:

- custom-built dew point traps and refrigerating circulator (2)
- Peltier-element (1, max. 2)
- mass flow controllers (6) and mass flow meter (1)
- gas pumps (4, max. 6)
- pressure sensors (5, max. 8)
- humidity sensors (2)
- magnet valves (24, max. 29)
- manual valves (4)
- Infrared gas analyzers (IRGA)

Some devices are directly connected to PC via standard interfaces: a Flow-BUS for the MFC's and the MFM, the IRGA, and the Refrigerating circulator are connected to RS232-ports each and the humidity sensors are connected to USB2 ports. As for the remaining devices the following data acquisitions (DAQ) modules and terminal blocks from National Instruments are used:

- SCXI-1100: 32 analog inputs (max. ± 10 V, 0 - 20 mA)
- SCXI-1124: 6 analog outputs (max. 0 - 10 V, 0 – 20 mV)
- SCXI-1163: 32 digital outputs connected to relays (0 - 24 V)
- TBX-1303: terminal block for 32 analog in (max. ± 10 V), additional shielding connector

The pressure sensors are hooked up with the TBX-1303 which is coupled to the SCXI-1100. The gas pumps will be actuated with the SCXI-1124 module. For both applications shielded cable has been used. All magnet valves are connected to the relays. They are contained in a relay-box, which was already in place. Just the electrical connections have been renewed partially with simple twin cable. 3/2-way valves with by-pass function (MV 1.1 + 1.2; MV 2.1 + 2.2; MV 8.1 + 8.2) and valves, which always switch simultaneously (V 5.3a + b; V 5.4a + b; V 9.2a + b; V 9.3a + b), share one digital output, respectively.

4.1. Collocation of the instruments

This section will describe the collocation of all devices in the 19-inch rack and the reasons for this alignment. In order to have a better overview and thus a simpler planning, the rack was subdivided into "levels", where the equipment is put together into groups (Figure 13, left). Within the realms of possibility, it was taken care, that there are as little changes between levels as possible. Furthermore the distance between the devices should be short and device types should be close together. The changes from one level to another based on the gas flow can be followed up with the help of complete flow chart in the attachments. The devices of one level are again grouped in color boxes according to those colors in the rack (Figure 13, left side).

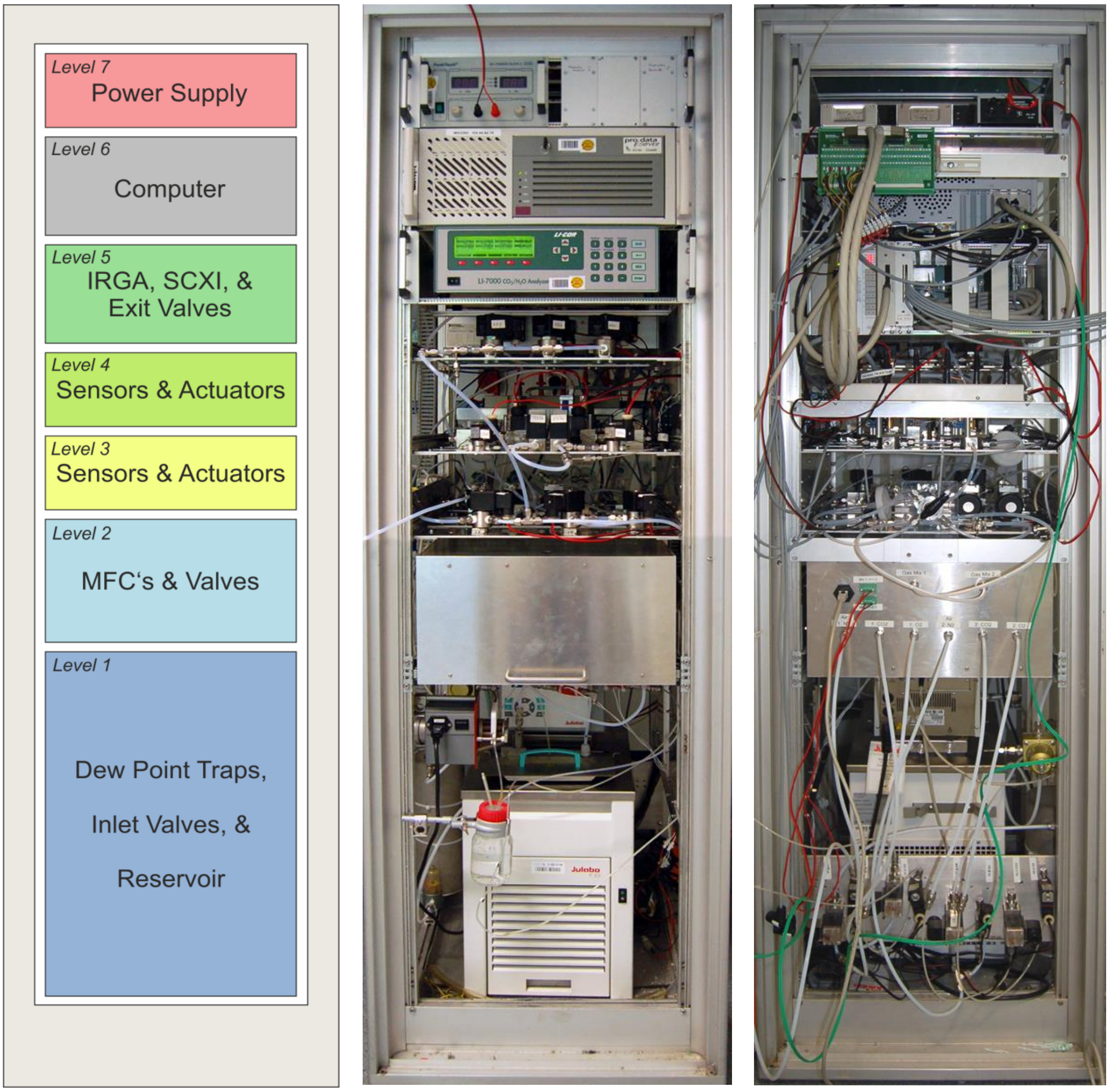


Figure 13: 19-inch rack design: left: basic "level" structure, middle: front view, right: rear view

The first level includes all four dew point traps, a gas reservoir, and the inlet valves (V 1.1 - V 1.4, V 2.1 - V 2.4). The reservoir is a hollow metal cylinder, which is used to homogenize the gas stream coming from the gas generator (CO_2RP140 Domnick-Hunter, Germany, with compressor OF302 JUN-AIR, Germany). It is attached on the left side. The valves are grouped on a metal plate, which is mounted on the rear side, where they can directly be connected to the gas supply lines. The dew point traps will be discussed separately (section 4.2 and 4.3).

The second level consists of the mass flow controllers (MFC's) and the remaining valves of the Gas Mixing (MV 1.1 + 1.2, MV 2.1 + 2.2).

Level 3 and 4 comprise all remaining parts of the flow chart system (discussed in section 4.6) except for the Infrared Gas Analyzer and some parts of the Gas Exit, which did not fit into these levels anymore. The height of the levels is defined by the levels (5, 6, and 7) above, the size of which cannot be changed.

The Infrared Gas Analyzer (IRGA) and the SCXI interfaces were positioned back to back in Level 5, where the IRGA is in the front and the Interface on the rear side. The IRGA is a Li-7000 from LICOR (Germany). It has 6 mm Swagelok fittings on its backside, where the gas lines will be connected. Because the SCXI-frame is a little bit higher than the IRGA, the space under the IRGA has been used to house the Exit-Valves and the absorbers of the Gas Exit.

The computer was placed directly above the IRGA and SCXI allowing a good connection to the devices and the SCXI. In front of the backside of the computer, the TBX-1303 for the analog inputs is fixed to a rail.

On Top (Level 7) four power supplies and two temperature controllers for the Peltier-elements are mounted. A power distribution board for six pairs of banana jacks (MC Power, NVT-620, max. 20 A, Germany) is connected to the main power supply (24 V) and positioned on Level 4 on the rear side, close to all devices.

The Relay-Box, which controls all valves, is attached on the upper right side of the rack and range from level 3 to level 7. Cable ducts complete the setup.

Additional space for the radioactive tracer application module is not available at the moment. Only a second Peltier-element has been implemented already and ports for the respective valves and pressure sensors of this module are available.

4.2. Setup of the humidification of the gas mixing modules

The refrigerating circulator takes up most of the space inside the rack and dominates the height of the level 1. Because it is very heavy and bulky, it stands on the bottom of the rack. All the other equipment for humidification and dehumidification is attached around it. Basically the setup of the humidification and the dew point trap for Gas Mixing and Conditioning follows the one in Figure 12 on the right side. The gas stream for humidification is coming from the level above, passing the water inlets and the heater mats first. Behind the heater mats, the gas is directly guided to the custom-built dew point traps, which are aligned in parallel in the water bath. Condensed water is again removed by the peristaltic pump. The pump is fixed close to it in the left front, so it can easily be reached for switching it on and off. Furthermore it can hold four tubing lines at once, which allows also the removal of water inside the Peltier-elements on demand. As water reservoir a glass bottle with open lid and silicone septum has been used. The silicone septum has four holes for the 3 mm water tubing. If one dew point trap is not in use, the water tubing should be removed from the peristaltic pump. If the water tubing of one unit is removed completely, two of the holes in the septum can also be closed by a short piece of tubing.

4.3. Peltier-element and temperature controllers

For the dew point trap of the main cycle (DP 8.1), no heating unit is necessary. It has been removed according to Figure 14 (left side). The water, which accumulates inside the dew point trap, still has to be removed. It can only be collected in a water reservoir, if there is an opening for pressure release. In this part of the system, exchange with the environment must be avoided under any circumstances, because of an error in measurement and the exposure of the environment if radioactive gas is released. Back-feeding the water tubing into the Peltier-element through all fittings solves this problem. In this way the water, which flows into the Peltier-element helps to remove single drops in a continuous flow. Each Peltier-element has an own water reservoir. As mentioned before, the peristaltic pump can also be used for the Peltier elements. To empty the Peltier-element, the water tubing has to be detached; eventually the hose pump has to be utilized additionally.

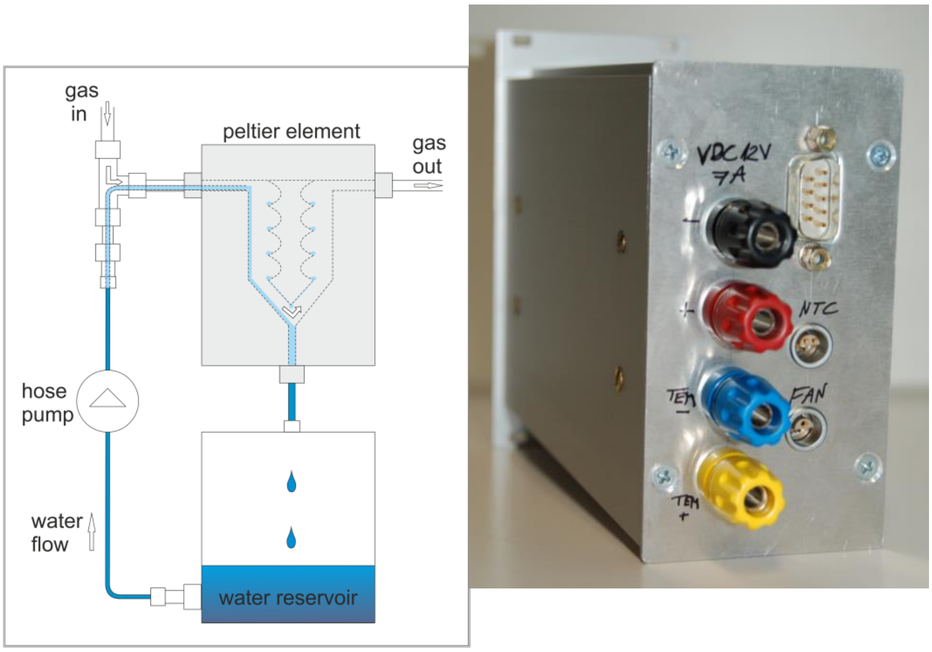


Figure 14: left: Peltier-element and cooling unit, right: housing for temperature controllers (rear side)

The Peltier-elements are attached on the lower right side of the rack, pointing outwards. In this way, fans of the Peltier-elements can blow the warm air from the heat sinks away from the rack. In consideration of the flow chart order, it would be good to position the Peltier-elements in intermediate height. But because of the risk of condensation of water in the line, it is advisable to locate them deep down.

As it was already indicated in chapter 3.3, temperature controllers (PR-59, Supercool, Sweden) replace the controllable power supplies, which have been used in first place. Two of these expensive power supplies would have been needed to operate the Peltier-element, resulting in a loss of one complete level of the rack just for the operation of the Peltier-elements. Another reason is that the temperature controllers and fixed-voltage power supplies (dedicated for the Peltier-elements and much smaller) were already in place (though never been used). In order to have a feedback signal, a NTC temperature sensor (delivered with the controllers) was placed in the cooling block of the dew point trap. For this purpose, a 15 mm deep hole had been drilled into the cooling block on the upper side providing a tight fit of the NTC sensor. Heat sink paste ensures a quick energy transport towards the sensor. The temperature controllers also allow the controlled power supply of the fans, so no additional power supply is required.

Because the temperature controllers are just delivered as boards on an angular heat sink, a casing is required to protect the electronics from dust and to avoid any contact with high (deadly) currents. A slide-in module for a 19-inch frame (3 RU) is taken as casing for each (see Figure 14, right). The connections were placed on the rear side in order to have a short way to the power supplies and the cable ducts. Because of the high currents, for the connections of the power supply and the Peltier-elements banana jacks in different colors were used. The respective cables and plugs have the same color code to avoid polarity reversal as far as possible. The NTC sensor and fan connections are realized with reversal protected circular plug-in connectors. Furthermore, the boards will be controlled remotely by the PC via RS232, for which a respective socket has also been placed on the rear side of each slide-in module. The modules are mounted on the front right side of level 7.

4.4. Mass flow controller drawer

The mass flow meter and mass flow controllers (Bronkhorst-Mättig, Germany) were taken from a previous system. Each mass flow controller (MFC) provides a defined mass flow rate of a specific gas, based on a reference input. The maximum flow rates are 500 mL/min for Air and N_2, 100 mL/min for O_2, and 5 mL/min for CO_2. The MFC’s and the MFM are measuring thermally and thus are sensitive to draft. For this reason the MFC’s have to be contained in a closed box. In order to have easy access it was decided to use a drawer as casing, the lid of which can be removed. Since there was no such drawer with the height and length required, a drawer has been planned and built in the on-site workshop. The construction is such way that every wall can be removed to drill holes into it for connections. Every connection is pluggable, so that the drawer can be taken out of the rack, completely. For the gas inlets and outlets, bulkhead couplings have been placed on the backside wall of the drawer, so they are close to the gas inlet valves and gas supply in the lab (Figure 15, left).

Inside of the drawer, the two sets of mass flow controllers are arranged in parallel and attached to sheets of aluminum (Figure 15, right). They are just fixed to the bottom with Pritt Multifix (Germany). They are hooked up to RJ-45 female connectors at the inner right side, which are again connected to one socket at the back wall. In order to mix the low-flow gas with the high flow-gas behind the MFC’s, the manufacturer provides a special T-piece (HI-TEC Gasmischer 7.01.446, Bronkhorst Mättig, Germany). It serves as inject nozzle and prevents the high-flow gas (CO_2-free Air + N_2) from entering the low-flow gas line (CO_2).

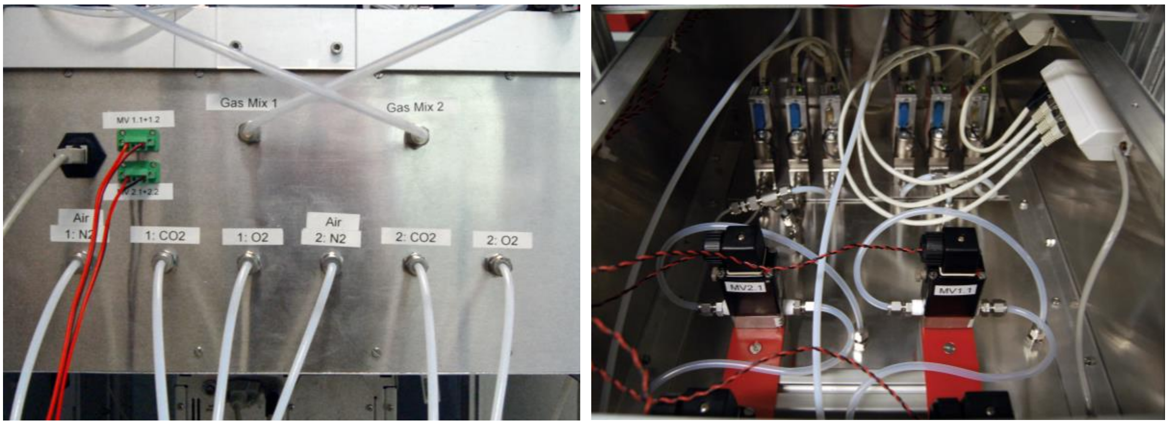


Figure 15: MFC drawer, left: rear side with connection, right: view inside with valves and MFC's

To avoid long gas lines, there should be a direct line to the humidification unit and the dew point trap. For this, the multi-valves MV 1.1, 1.2, 2.1, and 2.2 also have been placed into the drawer. The valves have been joined on a frame, so that they are lifted for about 5 cm (technical drawing in attachment). This makes the connection to bulkhead couplings in the bottom easier. The frame is again fixed with Pritt Multifix. The valves are electrically hooked up with a socket at the rear side of the drawer.

4.5. Ball valve actuators

For the 4/2-way valves MV 5.1 and 5.2, it was decided to take 4/2-way valves from Swagelok (SS-43YF2-125). The principle flow pattern is demonstrated in Figure 16 on the left side(Swagelok, Catalog 2013): Two of the four openings are connected each. By switching the valve by 90°C, the openings are connected across. The valves provide high tightness and are small at the same time. Furthermore they can be manufactured with a wider hole of 1/8 inch in the ball (3.18 mm), which causes a smaller drop in pressure at this point than a thinner hole. Unfortunately, the manufacturer does not offer an electrical actuator for the valve and pneumatic actuators occupy a lot of space and require two additional valves, which are connected to the compressed-air line of the lab. (As it is known, this will be needed for the gas generator.) For this reason, it was chosen to build custom-designed actuators with an electric motor and a mounting. The idea is based on a previous setup, which could not be implemented here. On a digital out signal, the valve conducts a 90° rotation. Limit switches indicate the right position of the valve and stop the motor. There were four basic requirements for the choice of the electro motor: First, it has to perform the rotation in less than 1 s to minimize significant pressure increase inside the line. Second, it should have more than 1 Nm of linear momentum (about 0.4 Nm is needed to turn the valve). Third, the motor should be a brushless DC motor to allow for fast acceleration in both directions. And fourth, the actuator should be as small as possible. For the actuators, an EC45 flat motor (BL Y 50W KL 1WE A) with the gearbox GS45 (2 NM 3ST KL 18:1) from Maxon-Motor (Germany) is taken. A DEC 50/5 amplifier (Maxon-Motor) controls the electro motor. The motor-gear combination and the valve are joined together on a mounting (technical drawing see attachment), which was constructed in the on-site workshop. A coupling made of brass connects the motor shaft and the valve shaft. It is shaped in that way that it also works as indicator for the position and triggers the position switches (1050.5305, Marquardt micro switches, Germany). The whole setup can be seen in Figure 16 on the right.

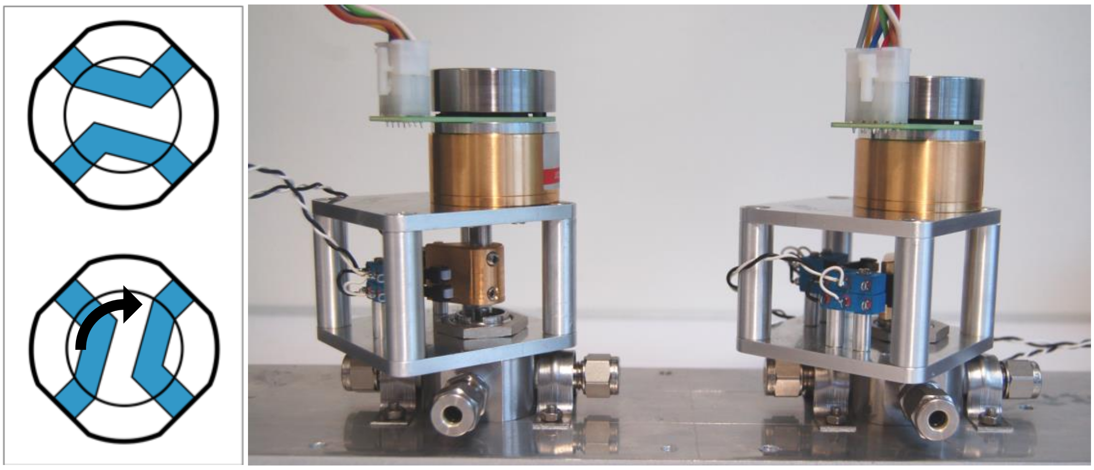


Figure 16: 4/2 way valves: left: flow pattern, right: valves with actuators and mounting

One valve plus actuator and mounting is about 12 cm in height and 7 cm in width. The switches are hooked up in such way that they are normally open (not triggered). In case of a failure of one switch, the motor would stop at a wrong position. For this reason, a second switch is connected in parallel for each position. The motor controllers are attached on the inner right wall of the rack. Because an independent unit has to check if curtain requirements are fulfilled, before a valve is switched, a microcontroller is needed for each setup. This microcontroller is realized with an Arduino board UNO (SMD-R3, Italy). They are contained in a special board case for Arduino's, which are attached left and right from the relay-box. There are directly connected with the digital outs inside of the relay-box (but without relay). The voltage supply (5 V) is also taken from the relay-box.

4.6. Sensors, pumps and valves of the Main Gas Cycle

As there is only very limited space for all remaining devices of the Main Gas Cycle, the arrangement of all of them in Level 3 and 4 (and partially 5) needs careful planning. As in the previous gas exchange system, the devices are mounted on 4 mm aluminum plates (11 cm in width) by screws. This allows for a modular setup and the tubing can be lead through the gaps of the plates. Four plates fit in level 3 and 4 each, but just two fit in level 5 due to the SCXI modules and the IRGA. During the planning phase all devices were grouped according to their function and position (e.g. pumps, pressure sensors, valves…) inside the gas cycle. This gives better orientation and helps to bundle electrical connections. It has also been taken care that flow ways are as short as possible and easy access is guaranteed for manual valves. In order to allow a flexible planning, often more footprints (screw hole patterns) have been prepared to attach the devices than necessary. Before the plates were fixed at their final positions, they have been moved in a quasi-setup to find a good arrangement. The final setup can be derived from Figure 17.

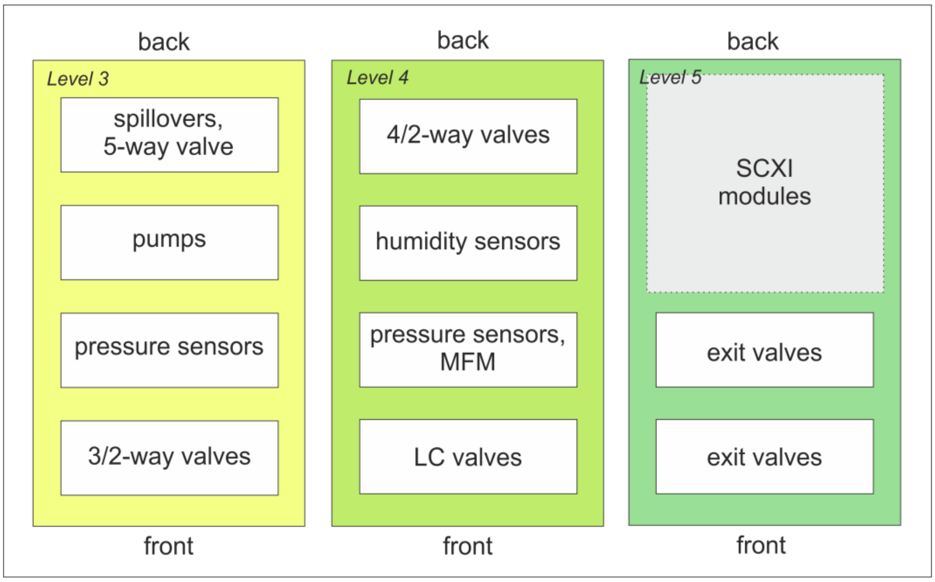


Figure 17: Arrangement of devices in level 3 and 4 (and partially 5)

The arrangement of the devices mostly follows flow way in the gas system. Starting at the rear side, the gas lines exit the MFC's' drawer and shall pass a pressure measuring point and a spillover before they pass the manual valve MV 4.1. The connection for spillover and pressure measuring branch (capillary) are combined in one part. Both parts are placed on the first plate in level 3 on the rear side. To have easy access to the manual 5-way valve it is mounted on a transparent Lucite plate on a rail perpendicular to the plate. So, the valve handle is pointing towards the back. The filter (Acro50 PN4003 1.0µm PTFE, Pall Corporation, Germany) is directly attached to the output of the valve.

The second plate in level 3 comprises all pumps, which are considered for this thesis. The gas pumps GP 4.1, 8.1, 9.1 are bigger in size than the remaining pumps and operate at 24 V with max. 5.0 L/min (NMP850, KNF Neuberger, Germany). The flow rate can be set by a control voltage between 0 and 5 V. Gas pump GP 4.2 (and the remaining pumps) delivers a flow rate of max. 1.3 L/min and operates at 12 V (NMP015B, KNF Neuberger, Germany). Because the 12 V pumps do not have a separate control input, an additional pulse width modulation is required in order to also connect them to the respective SCXI module. But the implementation of such is not part of this work. Nevertheless, holes for the addition of two small pumps have been prepared.

The connecting blocks for some of the pressure sensors (not the point of measurement inside the gas line) are attached to the next plate in level 3. These include the blocks for Pd 1.1 and 2.1, the capillaries of which end on the "spillover-plate", and Pd 8.1. The five available sensor blocks are completed by two additional blocks, which have been ordered at the workshop (technical drawing see attachment).

In level 4 the 4/2-way valves and their actuators have been mounted on the plate at the rear side for mainly three reasons: First, they are close to pumps GP 4.2 and 8.1; second, for maintenance they can be taken out easily; and third, the Application module for $^{11}CO_2$ can be easily attached. Coming from the ball valves, the gas lines towards and from the Leaf Chamber go in parallel. They first pass the humidity sensors, the blocks of which occupy the space of one plate. The next plate holds the pressure sensor blocks for Pd 5.1 (Leaf Chamber) and Pd 9.1 (Exit) and the mass flow meter MFM. As the MFM is also draft sensitive, a casing made of Lucite can be pulled over it. It has also been built in the on-site workshop (technical drawing see attachment).

On the last plate of level 4 the valves of the Leaf Chamber module are located. They are positioned in the front, to be able to easily connect a Leaf Chamber as well as the CO_2 trap. The Leaf Chamber implemented for the test runs is a custom-made Leaf Chamber with a cavity of 7 cm diameter and about 2 cm in height. It is sealed by two soft gaskets and silicone-based putty (Optosil R Plus, Heraeus Kulzer GmbH, Germany) in the mid plane, where the leaf would be placed (not part of this work). The gas streams into and out of the chamber through several small wholes (1.5 mm diameter). A picture of the Leaf Chamber is given below (Figure 18). The capillary for Pd 5.1 is directly connected to a pressure line, which enters the Leaf Chamber through an opening with 1 mm diameter. For more information it is referred to (Jahnke, 2001). In future, it might be advantageous to place the small gas pump GP 5.1 not on the “pumps”-plate, but close to the valves V 5.4a and b, so that easy connection of the CO_2 trap is still guaranteed.

Downstream the Leaf Chamber, passing again through 4/2-way multi valve MV 5.1, the gas is vented to the 3/2-way valves MV 8.1, 8.2, and 8.3 by pump GP 8.1. These valves are not located in level 4 due to limited space. A compromise has been found by using the last plate in level 3 in the front, so that they are positioned close to the gas pump and the Peltier-element DP 8.1. On the way to the IRGA and the Gas Exit module the gas passes the pressure measuring point for Pd 8.1.

As it was previously mentioned, the exit valves are placed together in level 5. They are mounted on two plates, where the CO_2 absorbers have not been implemented yet. They have just been bridged by tubing. Before the gas exits the system, it passes gas pump GP 9.1 on level 3.

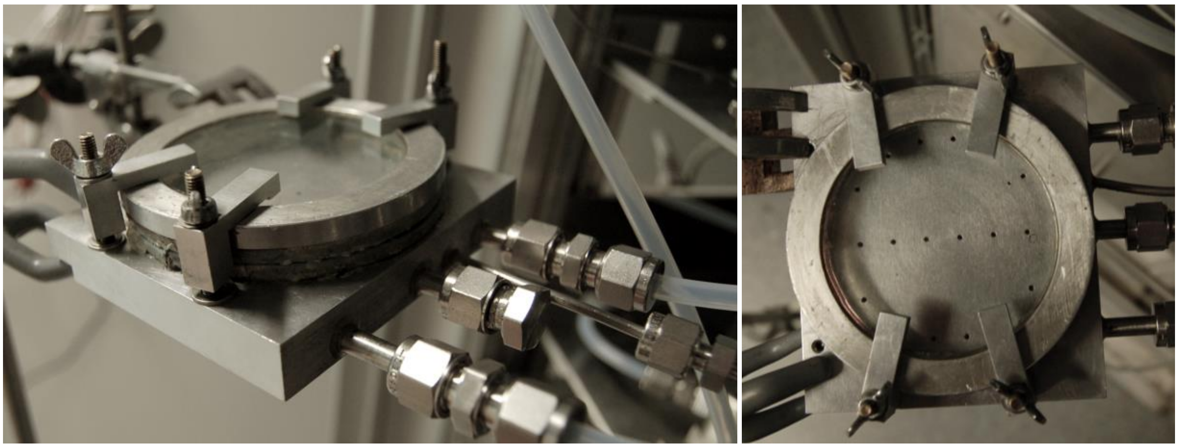


Figure 18: Leaf Chamber, left: gaskets and connections, right: top view with wholes for gas application

4.7. Dimensioning of the power supplies

Basically, three groups of low voltage are required: 5 V for the Arduino’s, 12 V for the small pumps and the Peltier-elements, and 24 V for all remaining devices, which do not have a power converter for 230 V/50 Hz. These are the pressure sensors, the bigger pumps, the MFC’s (and MFM), the relay-box, and the motor controllers. The 5 V for the Arduino’s are taken from the relay and will not be considered any further. In order to keep the costs low, it was decided to take individual fixed voltage power supplies for each of the voltage groups. Furthermore, one power supply dedicated for the use with the temperature controllers of the Peltier-elements was already in place and could be simply completed by another one (MW SP-100-12, 12 V/8.5 A, MeanWell, Netherlands). In order to determine the power consumption of the 24 V power supplies, the maximum power of the devices, have been noted in the table, containing all devices, and were summed up (in attachment). It was decided to take a fixed voltage supply from Elektro-Automatik 24 V with max. 10.5 A (EA PS524-11T, Elektro-Automatik, Germany). It is a small device and is offered with banana jacks.

All power supplies with fixed voltage are placed one the rear side of the rack. Left from the 24 V power supply, there are the two 12 V power supplies, one for each temperature controller.

On the front left side, a controllable power supply (DC 2250, Peak Tech, Germany) with a range of 0-32 V and max. 5 A is fixed. It was already in place and might be useful for any time later, like for temporary tests or for connecting further devices, e.g. the small pumps (12 V).

5. Software implementation

As was already mentioned previously, all devices are controlled remotely with the help of the PC software LabVIEW (National Instruments). The SCXI-modules operate with the driver software NI-DAQmx (National Instruments) and are already implemented in the programming interface. Furthermore, LabVIEW provides digital controllers, enabling the controlling of mass flow rate, pressure and dew points. Generally, the program structure has to fulfill certain criteria:

- All programs or program parts of the gas exchange system should be organized in a project structure. This includes programs, which have been used to address and test hardware independently from the remaining devices, programs, which are used as functions within the main program (SubVIs), specially defined control objects or libraries.
- As the gas exchange system is not completed yet and e.g. the Application module has to be implemented later, the program should allow for the simple addition of program parts.
- The user interface should be clearly structured to provide good overview. The handling of the program should be intuitive.
- The adjustment of the bath temperature of the refrigerating circulator to the set point (DP 1.1 and 2.1) can take up to 20 minutes. So it should be possible to configure the refrigerating circulator with a separate program independently from the main program. But it should also be possible to make changes about the configuration with the main program.
- As with the Peltier-element, the adjustment of the temperature can also take a couple of minutes. But an independent configuration is not required, since the reference is taken from the IRGA, which can only be derived if the main program is running.
- Acquired data should be saved continuously to file on demand, with a rate to be specified by the user. Data analysis can be done separately with other programs/routines (after the measurement). But online calculations of dependent parameters, such as CO_2 respiration, should be possible.

5.1. User interface (Front Panel)

The User Interface of LabVIEW is called the Front Panel. Due to the variety of switches and input and output windows (parameters) and the problem of keeping track of them, it was not advisable to locate them on a single page on the Front Panel. Hence, it was decided to divide them into sections similar to the gas flow charts and organize them with the help of tabulators (tabs). To follow up the gas flow, the tabulators are therefore named Gas Mixing 1, Gas Mixing 2, Leaf Chamber, C-11 Application and Gas Exit. The Main Gas Cycle tab is called JuGAS after the name of the old system: “Juelich Gas exchange and Application System”. Two additional tabs are included to summarize all current parameters and monitor them in graphs. Since the JuGAS tab is considered to be one of the most used tabulators, every other tab provides an additional button to turn back to the JuGAS tabulator. Above the tabs on the right a program stop button can be accessed from all tabs. Black arrows generally indicate the flow ways and directions.

Limiting the number of output windows to the most important parameters provides better overview in the first six tabs. The operator controls and indicators are labeled according to their functions in the gas flow charts to ensure easy identification. Input windows have a white background color; output windows are either light red (differential pressure) or light grey (remaining physical values).

Virtual LEDs with green and red color serve as indicators for the pressure status. A red LED shows that the pressure is not within a correct range. The Boolean operators for valves also look like their symbols in the gas flow charts. As an example a 3/2-way valve is given in Figure 19. The respective switch setting of the valve can be followed up by the light and dark green triangles, where light green indicates the connected valve openings (Figure 19, first column). If two valves are switched simultaneously, the second button has been replaced by an indicator with the same design of triangles (middle column). It should be noted here, that in case of the 4/2-way valves, the dark green triangles also symbolize a connection with each other.

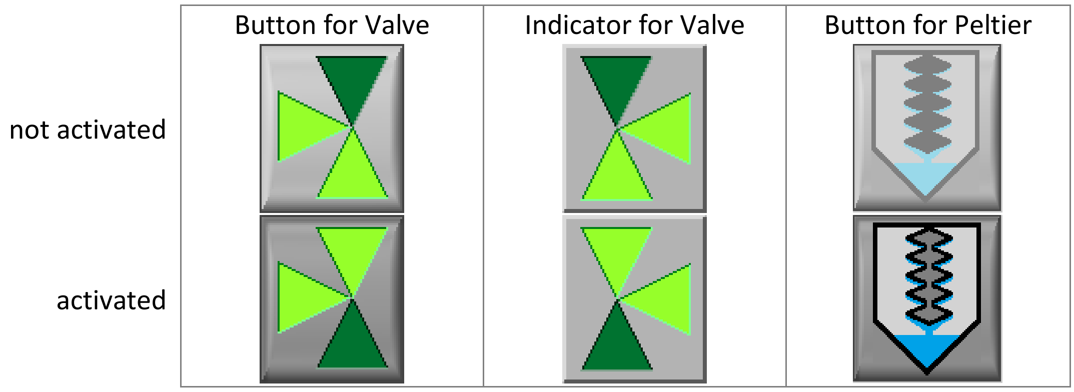


Figure 19: Button and indicator design for valves (3/2-way valve) and Peltier-element

The next sections will introduce the most important tabs. All other tabs are not illustrated here, but can be found in the attachment. Additionally the handling of the programs to configure the refrigerating circulator and to process the data will be explained.

5.1.1. The JuGAS tabulator

In the tabulator for the Main Gas Cycle (shown in Figure 20) other modules are shown as big boxes in the same colors, which have been used in the gas flow charts. To allow an intuitive change to each of the respective tabs, the boxes work as buttons. As it was described before, the buttons for actuating the valves look like their flow patterns. The buttons for the multivalves MV 5.1 and MV 5.2 trigger a movement of the respective engine, which turns the 4/2-way valves (for further information it is referred to section 5.3).

For multivalve MV 8.2 an indicator is implemented instead of a button. The 5-way valve MV 4.1 is not controlled remotely, so a button is not required. But to have a visual reminder on the valve, a panel with a similar valve design has been implemented (no control or indicator).

In order to engage the Peltier-element as dew point trap DP 8.1 of the Main Gas Cycle, a button with special design has been implemented, showing the cooling block with its ribs (Figure 19, right column). If it is activated, it will set the temperature controller to the operating mode and return the current temperature of the NTC. The dew point reference input (set point) is taken from the reference cuvette (A) of the IRGA in °C; the process variable is taken from the measurement cuvette (B) of the IRGA in °C (not shown).

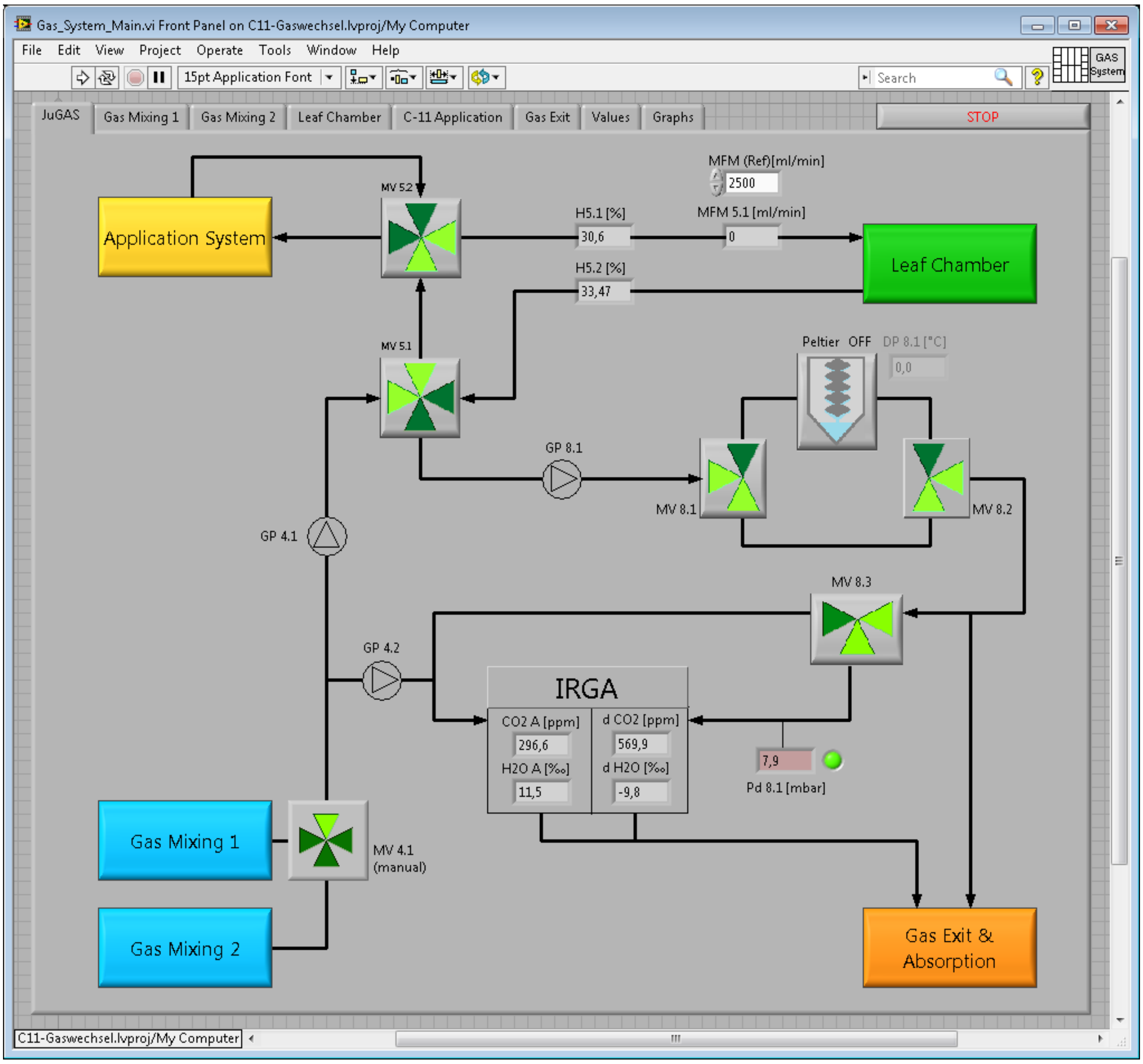


Figure 20: Front Panel of the main program showing the Main Gas Cycle tabulator

Just four values of the IRGA are displayed on this tab: the CO_2 and H_2O concentration of the reference cell A, and the respective differences of cell B with respect to A. A positive difference in concentration means, that the concentration in cell B is higher than in A. The LED next to it is set to alarm if the differential pressure exceeds $\pm$ 15 mbar.

The mass flow rate set point for the control loop (maintained by gas pump GP 4.1) is set in the white input window on the top (MFM (ref) [mL/min]). Underneath, the process variable can be monitored in the light grey output window (MFM 5.1[mL/min]). The values of the humidity sensors are reduced to the relative humidity (for temperature see Values tab).

5.1.2. The Gas Mixing tabulators

For each of the Gas Mixing modules one tab is provided. Here the tab is segmented into an upper and a lower part (Figure 21). The gas flow pattern is visualized in the lower part (incl. buttons for valves) and the controls for all set points are in the top, including an output window for the actual dew point of the refrigerating circulator. Here, the two different gas compositions modes are recalled, which can be used: Either CO_2-free air and CO_2 or N_2, O_2 and CO_2 are combined to obtain a defined gas mixture. Either selection is chosen by a red slide switch in the upper left tile. If the slide switch points towards “Air + CO2”, just CO_2 free air and CO_2 can be mixed. If the slide switch points towards “manual mixing”, the gas mixture can only be composed of N_2, O_2 and CO_2. Simultaneously with the selection of the gas composition mode, the respective valves are switched, which are generally disabled. For defining the MFC set points, pointer slides with additional digital display are used. They are disabled and greyed out, if the gas is part of the specified gas composition. If just one single gas is required (e.g. as it might be the case for matching or calibration purposes), the MFC set points of the gases, which are not needed, are to be set to zero. A button named “configure”, calls the Front Panel of another LabVIEW program, from which changes concerning the refrigerating and heating circulator can be made (will be discussed separately in section 5.1.5). This button is provided in each of the Gas Mixing modules, but is addressing the same device. However, the current dew point temperature is read continuously from the refrigerating and heating circulator and displayed in both Gas Mixing modules. An LED on the right side of the lower tile turns red, if the differential pressure is equal or lower than zero. In this case air from the surroundings will be sucked in causing a false measurement.

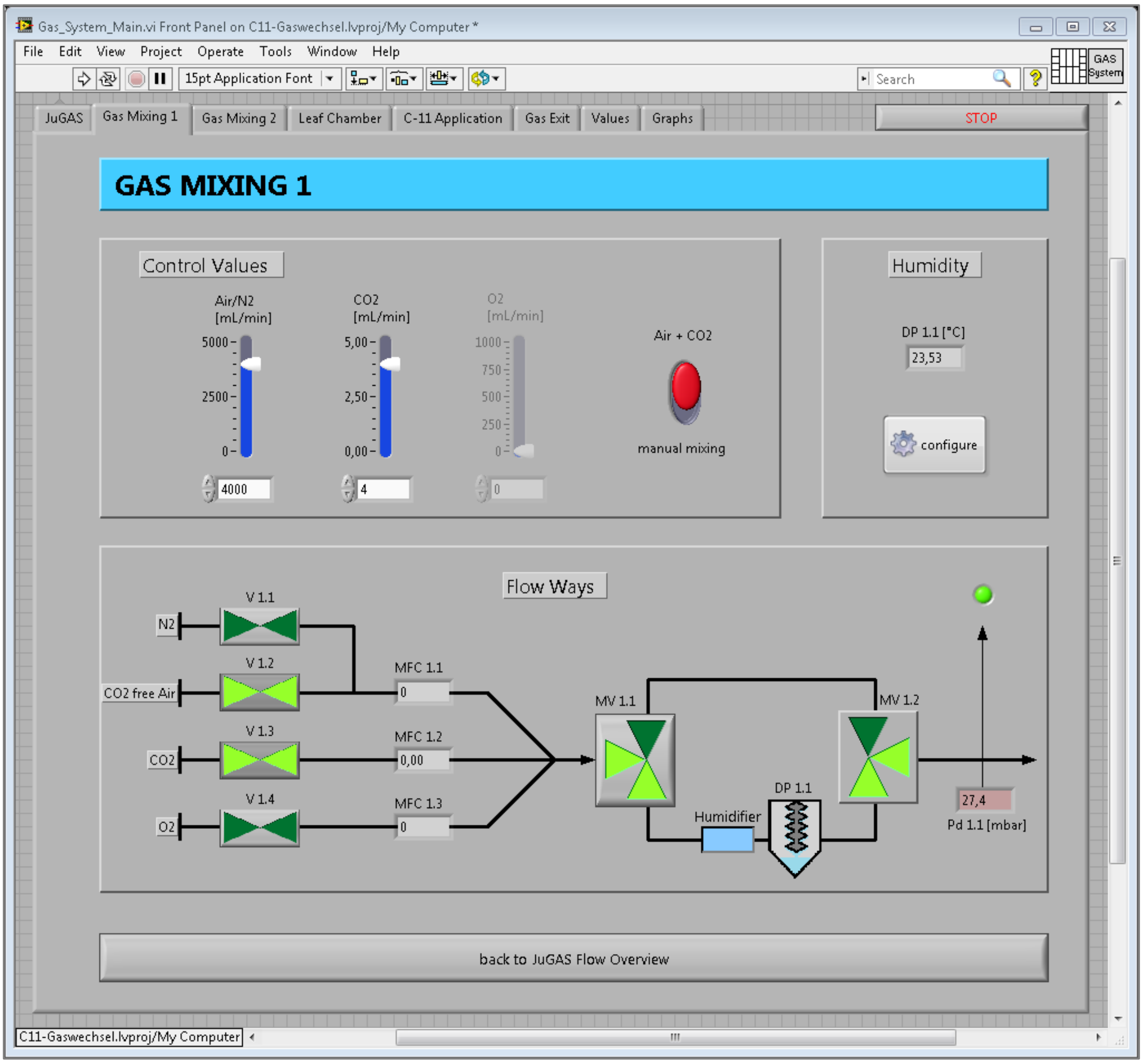


Figure 21: Front Panel of the main program showing the tabulator for the Gas Mixing and Conditioning module

5.1.3. The Values tabulator

In case, that values have to be viewed, which are not illustrated, it is referred to the Values tab (Figure 22). All numeric indicators are grouped in so-called "clusters" according to their physical properties. For each cluster a type definition has been added to allow easy changes afterwards (for more information see 5.2).

The LI-7000 (= IRGA) provides a long list of output parameters for CO_2 concentration and water vapor content for both cuvettes (and the difference) in different units. Here, eleven parameters have been selected to be sent from the device. The respective cluster is shown in the upper left corner. The CO_2 concentration is given in µmol/mol of gas (= ppm) with two digits of precision. The water vapor content is given in mmol/mol of gas (= ‰) with two digits of precision, too. Since the IRGA also delivers the humidity of the gas as dew point in units of °C, which can be directly taken as reference for the dew point trap DP 8.1, the dew point temperatures are also illustrated (one digit of precision). Additionally, relative temperature and absolute pressure are shown.

Below the IRGA cluster, the values of the humidity sensors are given. The sensors measure relative humidity in % and temperature in °C. Because output windows for temperature on the JuGAS tab will overburden the tab, these two values are shown just here.

To complete the group of parameters for concentrations and humidity, in the lower left corner the process values of the dew point traps (DP 1.1/2.1: Julabo, and DP 8.1: Peltier) are bundled to one cluster. The water bath temperature is given in °C with two digits of precision, the dew point temperature of the Peltier-element in °C with one digit of precision.

In the upper right corner the cluster for the MFC's and the MFM is located. It contains no further values than those, which are already included in tabs of Gas Mixing 1 and 2 (MFC's) and the Main Gas Cycle (MFM). The mass flow rate of N_2 and CO_2-free air and for the total flow (MFM 5.1) is given as integer, for O_2 as number with one digit of precision, and for CO_2 with two digits of precision.

All differential pressure values have been set together in a cluster, which is positioned below the MFC's and MFM cluster. If the program will be extended for the C-11 Application module, the additional pressure sensors have to be implemented into the cluster.

The last cluster on the right side comprises the control voltages from the pumps. This cluster is only required during test phases and will not be needed anymore, once the whole gas exchange system is set up. Currently, this cluster is defined as control since it might happen that the pumps have to be decoupled from the controllers. Later, the cluster can be quickly changed to an indicator cluster by two clicks. Variables have already been defined for all remaining pumps (small 12 V pumps), though they are not implemented yet. The reason for this is that the SCXI-1124 requires parameters for all analog output channels. So, they can be defined as constants (invisible) or controls, the latter of which has been chosen.

Controls to save all values have been placed on this tabulator due to limited space of the other tabs. All values are saved with a time interval, which is specified in seconds, in a file with a specified path as long as the button "Save data" is activated. If no path has been specified before, the user will be asked to define one. LabVIEW generally provides two types of writing data to a file: as binary or text data. The advantage of a binary data file type is that large amounts of data can be saved with high rates. This is possible also because the file, to which the data is written, cannot be accessed externally during the run of the program (if the button is activated). If less data amounts are to be saved, the data can be written to a text file and accessed during the measurement. Since it might be useful to make online calculations during the data acquisition to assess the outcome of a measurement, the text files were given priority. Furthermore, it is assumed that the amount of data is acceptable.

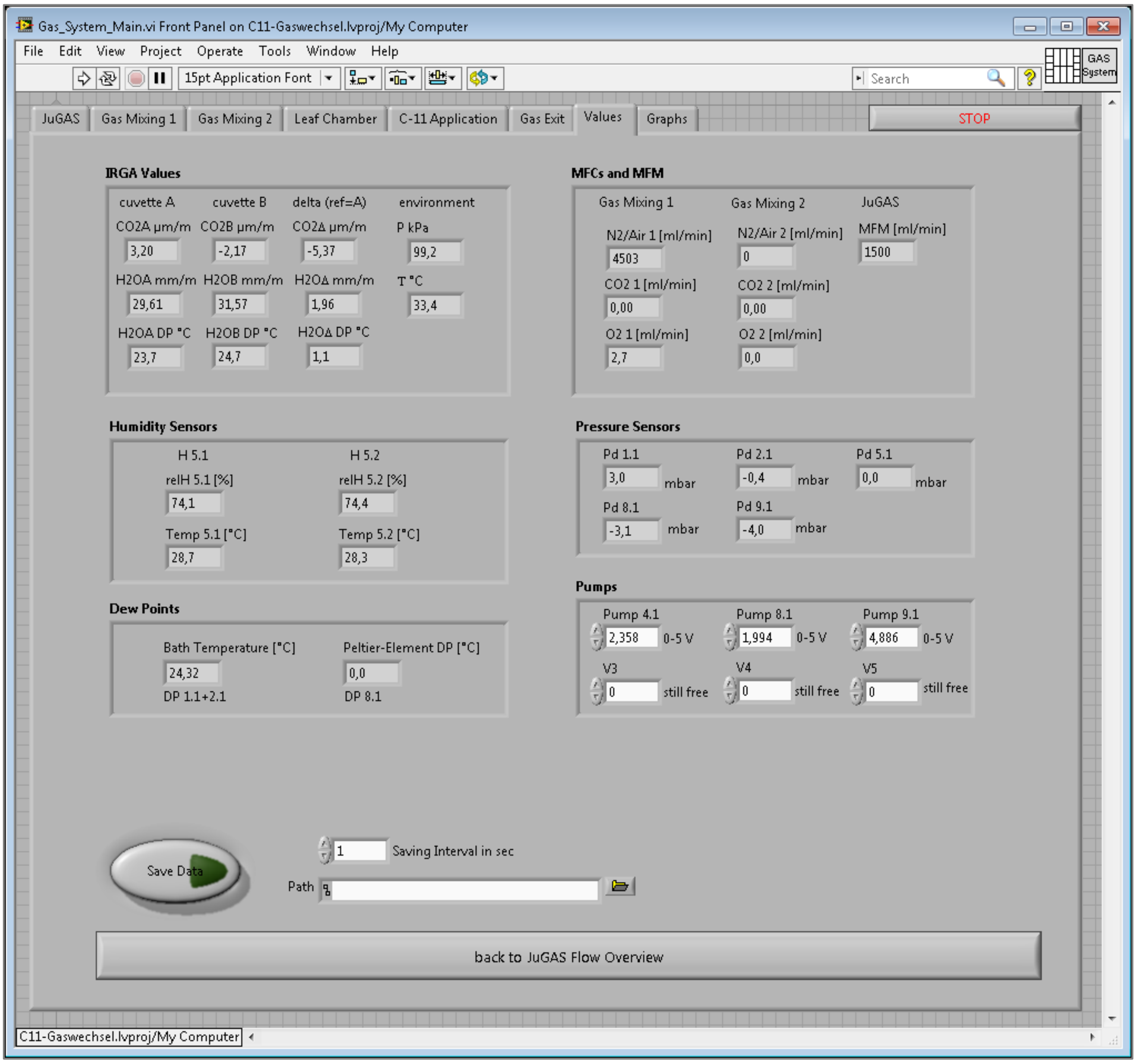


Figure 22: Front Panel of the main program showing the tabulator with all values

5.1.4. The Graphs tabulator

The Graphs tabulator (Figure 23) monitors all values, except for the pump voltages and some of the IRGA values (H2OB DP °C, H2OΔ DP °C, P kPa, T °C). Basically the values are again grouped according to their corresponding clusters. But this time, the values for dew point temperatures and humidity have been combined to one graph, so that there are four graphs in total. This was necessary due to the limited space on this tabulator. The graphs update according to the individual sample rate of the values. For this reason, the axis labels have been removed. Only the last graph has an axis label, showing the time in seconds as orientation. The humidity graph also includes the dew point temperature of cuvette A of the IRGA (DP ref). Since it is the set point for the Peltier-element

(DP 8.1), it is easier to compare both values if they are shown in one graph. The values of the MFC range from a few ml/min to maximum 5 L/min, so that the small concentrations cannot be seen properly. If the CO_2 concentration has to be displayed more detailed anyway, the scaling can be changed during the run of the program.

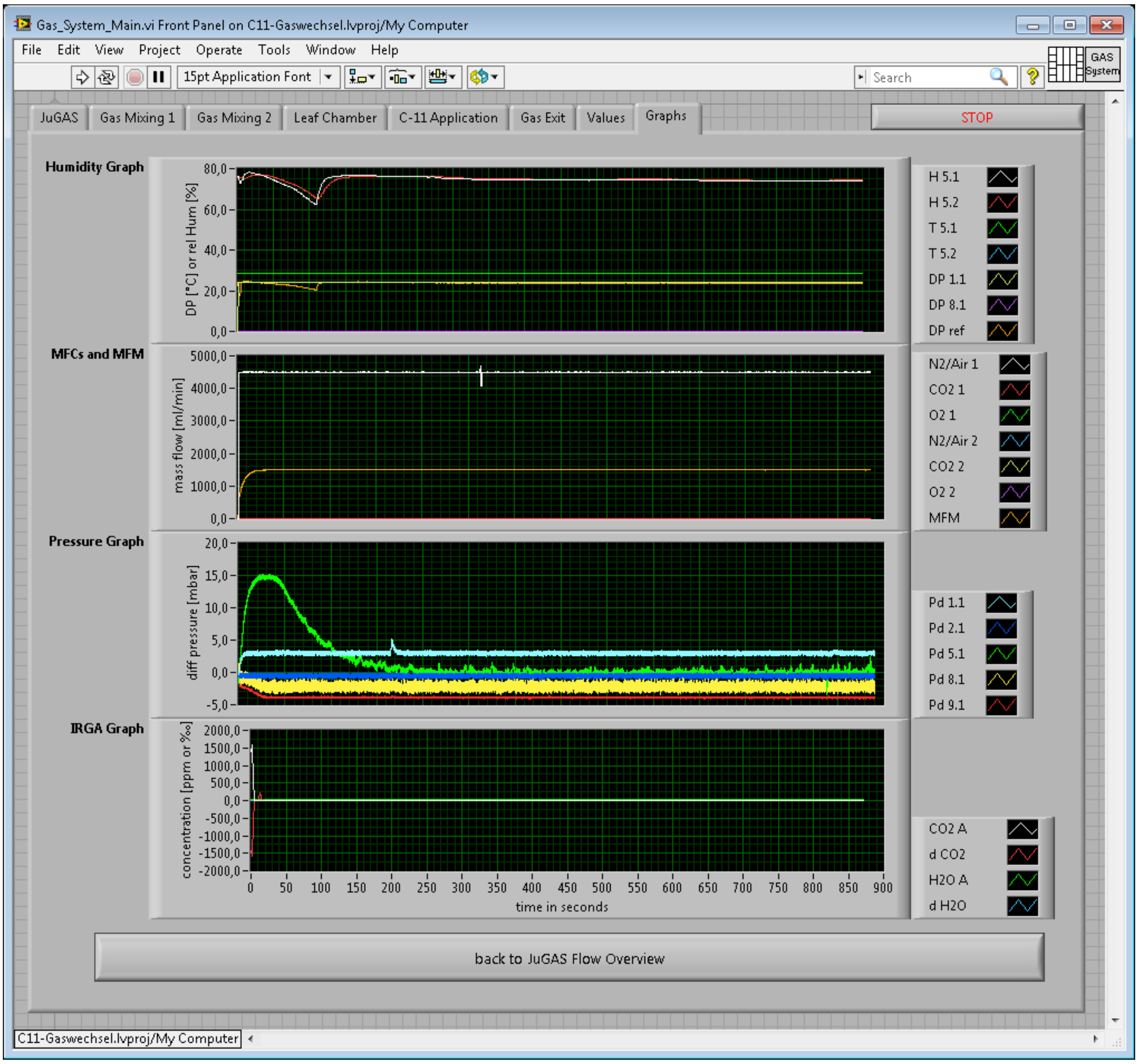


Figure 23: Front Panel of the main program showing the Graph tabulator

5.1.5. User Interface for Refrigerating circulator

The program to configure the refrigerating circulator before any measurement is the same program, which is called from the main program if the “configure” button is pressed. It is illustrated in Figure 24, while the program is running.

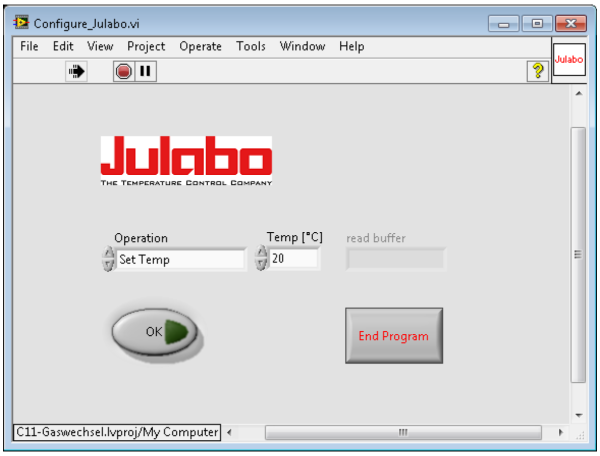


Figure 24: Front Panel to configure the refrigerating circulator

An operation can be chosen by the selector “operation”. Possible operations are: “Start” and “Stop”, “Set Temp”, “Evaluate Temp” and “Read Bath Temp”. The respective commands are sent to the refrigerating circulator by clicking the “OK” button. With the first two operations the refrigerating circulator will adjust to the set point or stop the control loop. A new set point is defined by selecting “Set Temp”, by which a numeric input window is activated, which is usually disabled and greyed-out. Here the value for the new reference temperature can be specified. By choosing “Evaluate Temp”, the current set point can be read or validated. Selecting “Read Bath Temp” derives the process temperature of the refrigerating circulator. Each answer from the circulator is displayed in the output window “read buffer”, which is activated for this purpose. After the configuration of the device has been finished, the program can be stopped with the “End Program” button.

5.1.6. Processing of the data

With respect to the functionality tests described in chapter 6, not all of the data is required for the evaluation or any other analysis. Furthermore, the data are saved without table headers in order to keep the Block Diagram small. Hence, it was found useful to create a program dedicated to data analysis, which reduces the data according to a selection and adds headers with the data types and their units. The Front Panel of the LabVIEW program can be seen in Figure 25. The program is applied when the measurement has been completed.

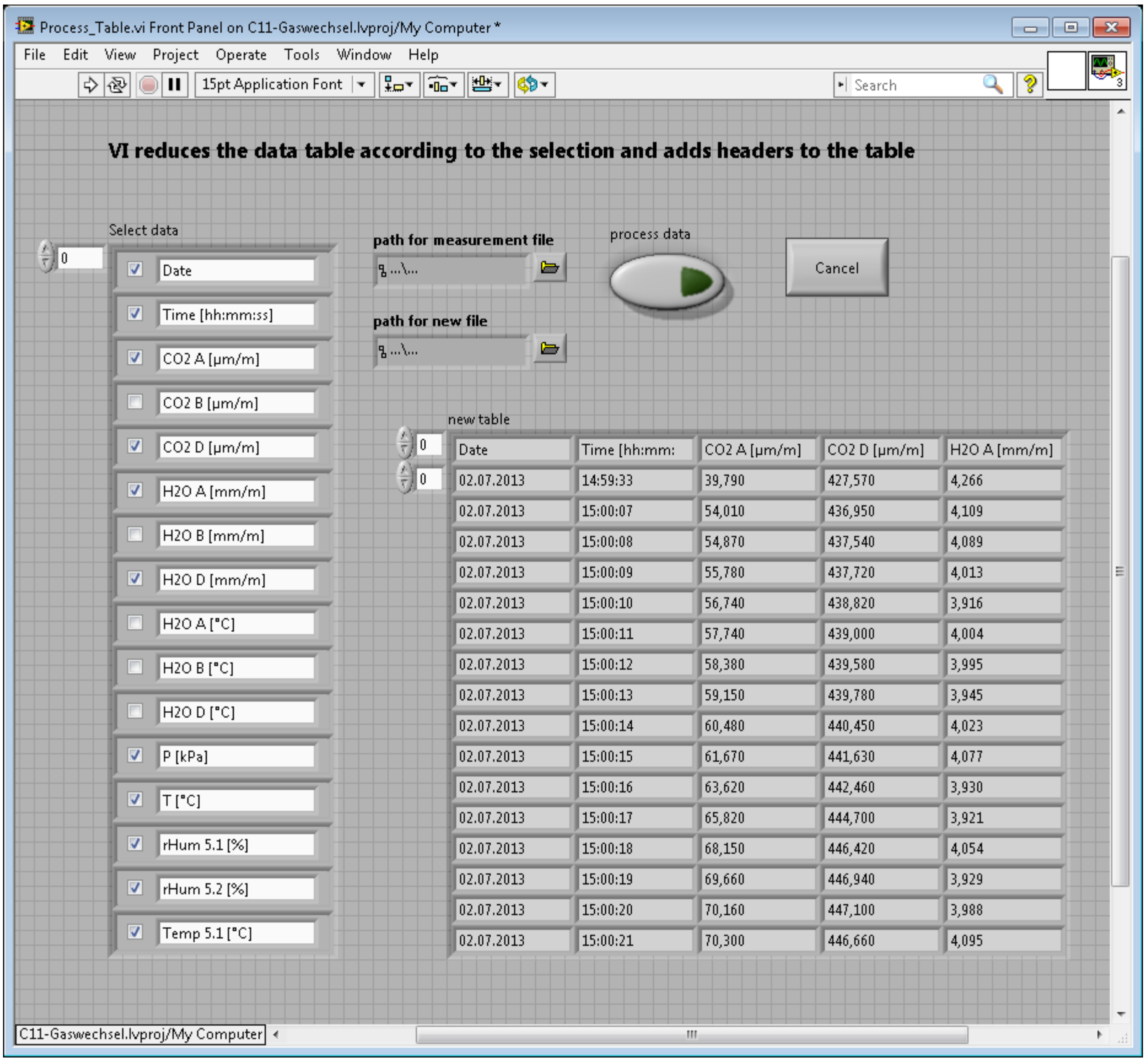


Figure 25: Front Panel of the program to reduce the data and to add headers

After the program “Process_Table.vi” has been started, the data required for data analysis can be selected by checking or unchecking the respective data type (window on the left side). By pressing the “process data” button, the user is prompted to specify the path of the measurement file and afterwards the path and name of the new file. The program stops automatically when finished. The table with the reduced data and the headers can be followed up in the big output window on the right side (new table).

5.2. Structure of the source code (Block Diagram)

In LabVIEW the source code is programmed graphically on an interface called Block Diagram. Since the handling of several parallel processes and communication with many devices simultaneously quickly leads to large and confusing program structures, many functions were summarized in so called “SubVIs”, which are separate programs within a main structure. Local variables were preferred instead of wired connections, if large distances had to be overcome or parameters from one function are used in another. As it was already mentioned in section 5.1.3, parameters were combined in clusters with type definitions to allow easy modification of the control. If a change needs to be made for a cluster, the change is made within the type definition and all dependencies are automatically updated. In order to apply a type definition, the cluster has to be saved as control.

To organize all dependent programs (= VI's), libraries and controls, a LabVIEW project was created, the tree diagram of which can be seen in Figure 26. The Project is divided mainly into two levels: The 1st level comprises the Gas_System_Main.vi, which is the program with the tabulators introduced in chapters 5.1.1 to 5.1.4, the Configure_Julabo.vi, which was introduced in 5.1.5, and the Process_Table.vi, which was introduced in 5.1.6. These three programs need to be found quickly by the operator of the system and are therefore separated from all other programs in the 2nd level. In the 2nd level all SubVIs or test VI's were inserted into a folder "SubVIs". The same is done with all controls (e.g. the special valve buttons and the clusters) in a folder "controls", respectively.

The source code of the main program "Gas_System_Main.vi" is divided into three temporal main parts with the help of a stacked sequence.

In the first step all variables are initialized and all ports necessary for the communication with devices are defined. The buttons for switching the valves are disabled. Furthermore, the temperature controllers for the Peltier-elements are configured. The configuration includes the starting of the cooling fans and the setting of a controller type (POWER mode).

In the second step the program carries out its main task: The values are read from the devices and commands are sent to devices, the digital controllers are passed through, the Front Panel actions are processed, and the values are saved to a file. In order to acquire the data or send commands the communication with each device is conducted with several parallel while loops, to allow for different, device specific sampling rates. Each of the loops will be discussed in the following sections.

In the last step all operations necessary for the stop of the program are placed: the valves are set to their default state, the pumps, the temperature controller for the Peltier-element and the mass flow controllers are stopped. The control loop of the refrigerating circulator is only interrupted, if the user confirms the request in a dialog window, which shows up after pressing the Stop Button. Additionally, all ports are closed.

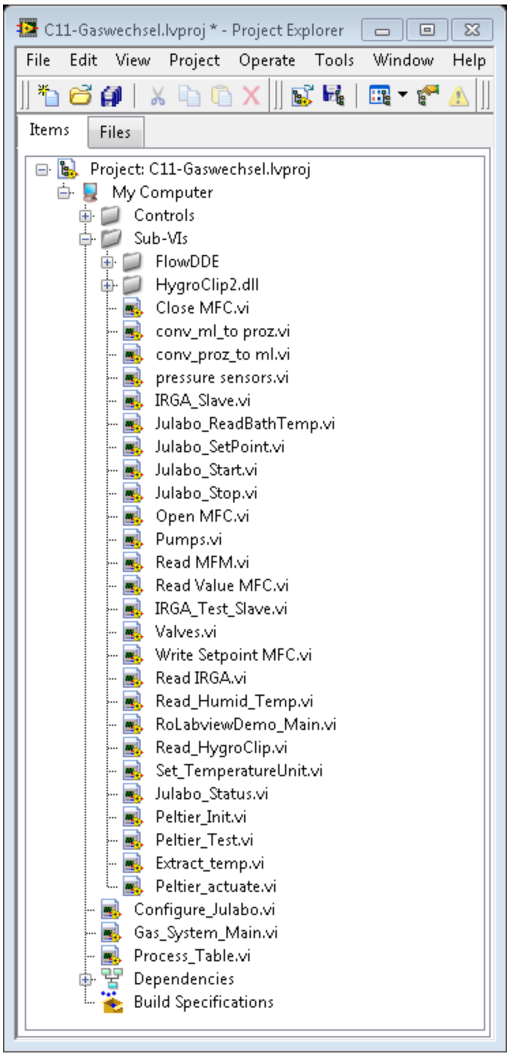


Figure 26: Tree Diagram of the LabVIEW project (update when Peltier ready)

Screenshots of all programs can be found in the attachments.

5.2.1. Structure of the Main Loop:

In this while loop the major functions of the program (like the reading of most of the buttons) are situated. In the upper part the Front Panel elements (valve buttons of the Gas Mixing tabs, mass flow controller input windows and output window for the Peltier-element temperature) are activated/inactivated/disabled & greyed-out/enabled with the help of property nodes depending on the switch state. The reading of the buttons for switching tabs is also located here. Underneath, the current differential pressures are read from a SubVI and are written to the Front Panel output windows. A comparison of the differential pressure values with the allowed limits yields a Boolean, which is written to the pressure status LEDs. For completion, it should be mentioned, that the LED for Pd 9.1 on the Gas Exit tab will turn red, if the differential pressure is greater or equal than zero. The pressure values are directly written to the graph.

In the left bottom, the controllers for controlling the mass flow rate and the differential pressures Pd 5.1 and Pd 9.1 were placed. If the button for multivalve MV 5.1 is activated, the gas exchange system is running in a closed cycle and the pumps GP 4.1 and GP 8.1 are decoupled from their references. In this case the parameter for the pumps should be fixed to the last value. For this reason, the controllers were placed in a case structure (= if-else-structure) where the selector is a local variable of MV 5.1. If the variable is false, the controllers will send a new voltage parameter to the pumps, else the last parameters are sent to the pumps. As for pump GP 9.1, it doesn't make sense to leave the pump running, if all exit valves (V 9.1, 9.2a + b, 9.3a + b) are closed. Here, the controller will be enabled (case structure true) if one of the exit valves are open. Otherwise the pump voltage will be set to zero. However, for test purposes, the selectors of all case structures will be temporarily disconnected from the local variables of the valve and connected to Boolean constants. On the right bottom of the while loop the current states of the valve buttons are sent to the relay-box. The while-loop is timed with 50 ms.

5.2.2. Loop for data acquisition from the humidity sensors

As was already mentioned in chapter 3.2, LabVIEW software for acquiring data from the Hygroclips is provided by the manufacturer. This program (including SubVIs and a library) is slightly modified and implemented as SubVI for each humidity sensor. The values are read every 100 ms and transferred to the respective Font Panel outputs.

5.2.3. Loop for handling of the mass flow controllers

This part of the program was already implemented in a program for the older gas exchange system and could be set up in the same way as before. It should be noted that a special software (Flow DDE) provided by the manufacturer must be running in order to communicate with the Flow Bus. In the while loop, two communication channels are opened: in the first channel, the reference values are written to the mass flow controllers, and in the second one, the process variables are read from the mass flow controllers and the mass flow meter. Two SubVIs handle the transformation of the values from % to mL/min and vice versa. The data are acquired with the same rate as in the main loop (= 50 ms), because the mass flow rate is one of the process variables of the control loops. The results are directly written to the Front Panel elements including the respective graph.

5.2.4. Loop for data acquisition from the IRGA

Here, it was possible to choose between two ways of acquiring data: either on request of the PC (data polling) or the IRGA sends data with fixed rate to the computer. Since setting up the first way is rather time consuming due to a poor description of the code in the manual, the latter way was chosen. For this reason the LI-7000 was set to send the 11 parameters with a fixed rate of 1s to the computer. Therefore, it was necessary to acquire the parameters also with exactly the same rate, which is realized with a timed while loop (trigger = 1 s). In contrast to normal while loops with a timer, these loops start counting the time, once a loop turn is started (not when they finish the loop).

A property node tests, if there are bytes at the serial port. If this is the case, they are read as string from the port and transformed into an array of double floats. The values are extracted according to their positions in the array, which is always fixed. The parameters are then written to their respective Front Panel elements. CO2 A, H2O A, d CO2, and d H2O are bundled to an array and displayed in the LICOR graph.

5.2.5. Loop to configure the refrigerating circulator

Since the communication between computer and the refrigerating circulator depends on the state of both "configure"-buttons of the Gas Mixing tabs, both Boolean controls are connected to an OR-function. The OR-function output is connected to the selector of a case structure: If none of the buttons is pressed, the current bath temperature is requested ("in_pv_00<CR>") and read from the device as string. This string is converted into a double float and written to the respective cluster and output windows of the Gas Mixing tabs. If either one of the buttons is pressed, two things happen in series: First, the SubVI "Configure_Julabo.vi" is called, resulting in an opening of the Front Panel, where the configuration operations can be chosen. The start command of the refrigerating circulator is "in_mode_05 1<CR>" and the stop command is "in_mode_05 0<CR>". The reference temperature (set point) is set by "out_sp_00XX.X<CR>" and evaluated by "in_sp_00<CR>", where XX.X is the set temperature. When the Configure_Julabo.vi is ended, the port is closed. So, in the second step, the port is opened again and both buttons are deactivated (false state). Because this part is not time-critical, the while loop has a slower repetition rate of 1 s.

5.2.6. Data communication with the temperature controller

The temperature controller can be used as stand-alone controller, which means that the feedback temperature is taken from the NTC in the cooling block. Because this is not necessarily the dew point temperature of the gas, it is desirable to take the process variably also from cell B of the IRGA. Since this requires controlling over LabVIEW, the controller will just be given an output variable between 0% and 100% (representing cooling power) and will be used as amplifier only. For this purpose the temperature controller will be run in the "POWER" mode. The controller output is referred to as "T_C limit".

For the communication with the temperature controller of the Peltier-element, a SubVI has been written, which allows the following operations: start the controller, stop the controller, write the set point, and read the current temperature of the cooling block from NTC sensor. The respective operations can be chosen by a text ring variable. The SubVI can be used several times in one loop turn.

If the button for the Peltier-element is activated, the temperature controller should be running until it is deactivated. Here, a problem occurs, because the temperature controller should be only started or stopped once, if the button state changes. But the button has switching behavior instead of latch behavior. So a way had to be found, to compare the current state of the button with its old state. This could be solved by introducing an integer variable ("stop/start index"), which either becomes "1" if the temperature controller was set to the running mode or "0" if the temperature controller was set to the stop mode.

Now, the while-loop for the communication with the controller is set up as follows: The Peltier-button (Boolean) is connected to the selector of a case structure: If the button is activated, the following functions are processed one after another: First, the temperature controller is started, if the start/stop index is "0", which means it is not running (additional case structure). Second, the start/stop index is set to "1" and the set point is transferred to the controller. Third, the current temperature of the NTC sensor is read and converted to a number by the help of the String_To_Number.vi. The result is displayed in the Front Panel elements. If the loop is conducted a second time, the temperature controller will not be started again because the comparison with the start/stop index will yield a false state.

If the Peltier-button is deactivated, the start/stop index is compared with a “1”: If the output is a true Boolean, first, the stop command is sent to the temperature controller, and second, zeros are written to the Front Panel output windows to indicate that the controller is not running. Furthermore, a “0” is written to the start/stop index, so that the controller is not stopped again. If the output of the comparison is a false Boolean, nothing happens.

The while-loop timing is adapted to the sampling rate of the IRGA (= 1 s).

5.2.7. Saving of the data

Saving of the data to a file is straight forward and organized in a timed loop. The saving interval time specified by the user (in seconds) is multiplied by 1000 to get the time in milliseconds and connected to the trigger input of the timed loop. If the safe-button is activated, the clusters are converted to arrays and converted to a string array. All string arrays or single strings are appended to a time stamp and saved to a spreadsheet file. Note, that for test purposes also the state of one button (e.g. multivalve MV 8.1) can be saved to the file.

Inside the timed loop (but not as part of the case structure), the reference dew point temperature “H2O A °C” is extracted from the IRGA cluster and written to humidity graph together with the relative humidity of H 5.1 and 5.2 and the temperature of the cooling block of the Peltier-element.

5.2.8. Processing of the data

This program will not be discussed in detail because it is just of minor importance. To process the data, first an array of clusters is defined (array name= “select data”). Each cluster consists of one header (String) for one specific data column and one check box (Boolean). The array is built with constants and written to an indicator, where the user is allowed to make his selection. If the user confirms his selection by pressing the “process data”-button, the data will be loaded and transformed into an array of the same type like the “select data”-array. Afterwards, both arrays are processed in a For-Loop according to the number of data columns: If the respective data is selected, then the column is extracted, the header is added and the column is written to a new array. This is saved to the new spreadsheet file.

5.3. Program for the Arduino Uno

The Arduino chips are loaded with the same program each, building the logic to enable and disable the motor controller of each motor for the 4/2-way valves. The motor controllers have three inputs important for turning the valves: /Disable, /Brake, and Direction. The backslash indicates an inverted input. The /Disable and /Brake input are connected together. This input is connected with pin 13 of the Arduino board. Direction is defined that way, that a GND signal means a movement counterclockwise (in the program called left) and a 5 V potential means turning clockwise (in the program called right). One digital out of the SCXI-1163 (without relay) is directly connected with the direction input of the controller and pin 8 of the Arduino board. The information of the current position is given by the end switches, which are called LES (left end switch) and RES (right end switch) in the following. They are connected to pin 3 and pin 2, respectively. All input pins of the Arduino (2, 3, and 8) are defined as using Pull-Up resistors so they are kept on 5 V if open. As soon as an end switch is pressed (closed), the potential of pins 2 or 3 drops to 0 V (GND). Similar to the Peltier-program part, a variable is defined to save the direction, into which the motor was turning the last time (= DIRold).

In order to not miss an event in a while loop with fixed iteration time, such as the reaching of an end switch, two Interrupt functions were introduced (for pin 2 and 3 each). The function performs a task as soon as specified condition occurs. Here, the Interrupt function disables the motor controller, as soon as either end switch is pressed. Since the end switches show bouncing behavior, it was necessary to trigger the function, if one of the input potentials drops to GND potential (FALLING).

The Interrupts are implemented in a normal loop function, which calls one of the Interrupt functions, if the motor is turned in the respective direction. Each case for turning left and right is handled separately and will be explained on the example of turning the valve counterclockwise: The user presses the button for turning one the 4/2 way valves and the pin for direction will change to GND. Inside the loop, an If-Structure now checks if the current direction (DIR) is left, AND if the old direction (DIRold) was right, AND if the left end switch is not pressed. In case of a true, the Interrupt function for the right end switch is disabled (due to bouncing of the switches), and the one for the left end switch is enabled. The motor is enabled and turns left. The “left”-direction is written to DIRold. The motor has performed a 90° turn and touches the left end switch. The Interrupt-function is called, which stops the motor.

Turning in the right direction is handled accordingly.

6. Evaluation of Functionality

The aim of this chapter is to prove the correct setup of the gas exchange system and the program, as well as to determine the application ranges of the system. As operating point for this specific Leaf Chamber a mass flow rate of 1500 ml/min is defined for later measurements. The tests include:

- Evaluation of the correct assignment of the interface controls (and channels) to the sensors and actuators. This is especially important for valves, pressure sensors and mass flow controller. This part will not be discussed here, since this is straightforward.
- The diameter of the spillovers has influence on the mass flow range and the differential pressure of the system. Here, it should be determined, how this influence can be minimized.
- The system should be applicable for several Leaf Chamber sizes, which means that the system should be able to operate at different mass flow rates. For this purpose, the range of the mass flow rate is to be determined and the control process for the mass flow rate should be optimized for the new setup. Further, the quality of the chosen control parameters is to be assessed.
- In the course of this, the control processes for the correct pressure status in the Leaf Chamber and at the Gas Exit are to be adapted. The differential pressure measured by Pd 5.1 should have a deviation within ± 1 mbar; the one measured by Pd 9.1 should be slightly negative (for different mass flow rates).
- The Gas Exchange System should enable measurements under different CO_2 concentrations. Hence, the adjustment range for low, atmospheric and elevated CO_2 concentration is to be determined with the IRGA.
- For quick changes of CO_2 concentrations and calibration purposes, dwell times for CO_2 should be estimated.
- The range of the initial dew point temperature of the gas provided by the refrigerating circulator is to be determined. The resulting humidity is to be monitored with all devices measuring humidity (humidity sensors and IRGA).
- Humidity resulting from transpiration of water vapor by a plant leaf inside the Leaf Chamber should be eliminated by dew point trap DP 8.1. Therefore, control parameters for controlling the Peltier-element temperature by the temperature controller are to be evaluated.

Before each point is discussed in more detail in the following sections, it should be mentioned that a test using the Gas Generator proved a malfunction: the CO_2 concentration of the CO_2-free air was 400-500 ppm. So, it is assumed that the gas is not absorbed at all. CO_2 concentration measurements will therefore be performed with N_2instead of air. The humidity of the dry CO_2-free air was determined to be 5 mmol/mol, which is still acceptable for short test measurements. However, for the proper working of the gas exchange system, a new gas generator has to be installed.

6.1. Adjustment of the spillovers

As it was described in section 2.2, abundant gas of the initial gas mixture is to be released at the spillovers in the Gas Mixing and Conditioning parts. To ensure that no air is sucked in, the differential pressure sensors Pd 1.1 and Pd 2.1 should have positive values. In order to maintain this condition, the initial mass flow provided by the mass flow controllers should be higher than the mass flow rate measured by MFM 5.1. The exact difference between initial mass flow and Main Gas Cycle mass flow depends on the diameter of the spillovers. Spillovers with small diameters cause high flow resistance and can lead to pressure increase in the system, which is difficult to compensate. Additionally, there is an offset mass flow in the system without any pump running, which can make measurements in the low flow rate region difficult or even impossible. The bigger the diameter, the more gas can leave or enter the spillover tube. At lower initial mass flow rates, there's a higher risk of sucking in air from outside. Consequently, the initial mass flow rate has to be set much higher than the Main Gas Cycle mass flow, resulting in high gas consumption. For this reason it is crucial to prove that the spillover diameter is chosen correctly.

For this test, the mass flow controller value MFC 1.1 of CO_2-free air or N_2 has been increased stepwise by 500 ml/min from 0 to max 5000 ml/min. The gas was taken from the gas generator. The pumps were not operated. All valves were set at default switch mode except for valve V9.1 being open. The mass flow rate MFM 5.1 of the Main Gas Cycle and the differential pressures Pd 1.1 and Pd 5.1 were measured as a function of the initial mass flow rate provided by the mass flow controller of the Gas Mixing 1 module. The results are given in the graph below (Figure 27). The mass flow rate MFM 5.1 is scaled on the primary y-axis and is plotted in red; the differential pressures are scaled on the secondary y-axis and are plotted in blue (Pd 1.1) and green (Pd 5.1). The filled data points in the upper part of the diagram show the influence of the spillover with 1 mm diameter, which was implemented in first place. The resulting mass flow rate MFM 5.1 was about 2 to 3 times less than the initial mass flow rate, leaving not much operating range for the pumps. Even more important is the pressure increase inside the system: at maximum MFC value of 5000 mL/min, the differential pressure at Pd 1.1 was already out of scale (more than + 70 mbar) and the differential pressure at Pd 5.1 still showed + 35 mbar, which is difficult or impossible to compensate by the pumps. In addition, it is not accounted for the pressure increase inside of the Main Gas Cycle if the pumps are operating. This is a clear indicator that spillover diameter has been chosen too small.

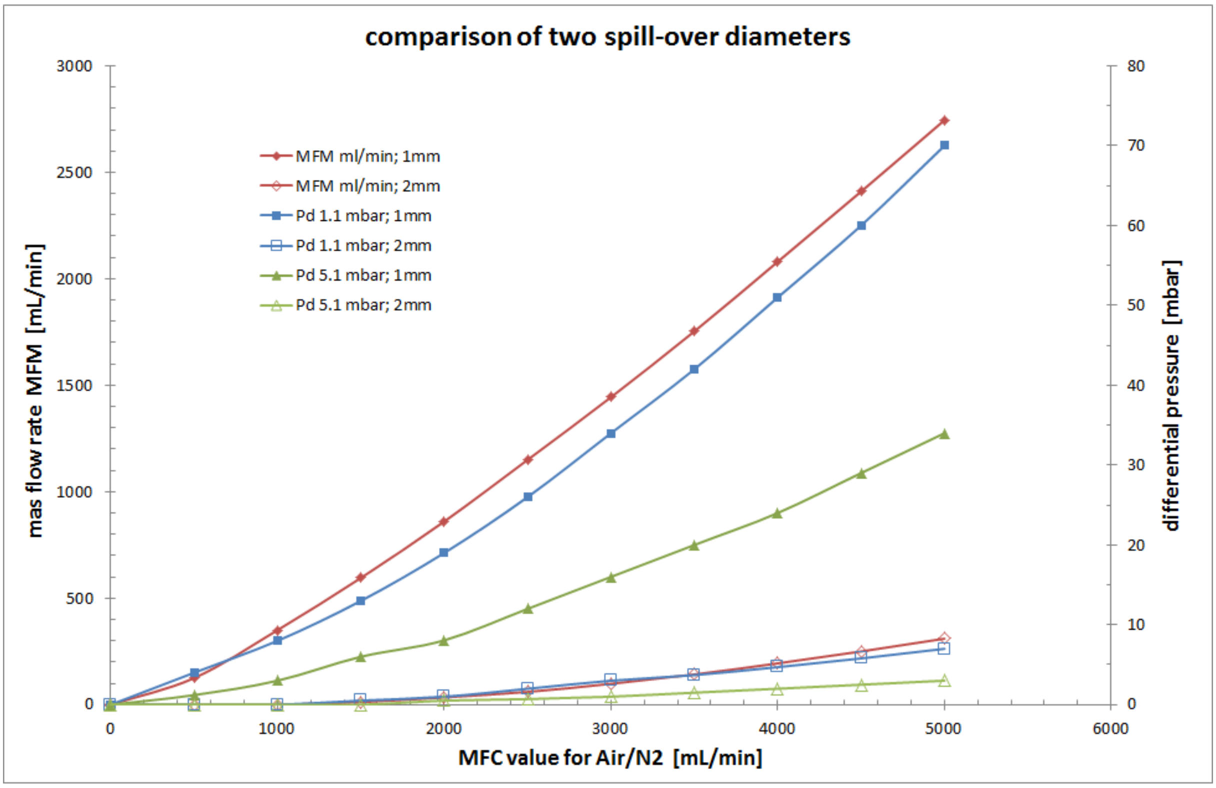


Figure 27: mass flow rate MFM 5.1 and differential pressure Pd 1.1 and Pd 5.1 in dependence of mass flow controller process variable while no pump is operating: comparison of two spillover diameters: 1 and 2 mm

As a matter of fact, the 1 mm open diameter tubes serving as spillovers were replaced by 2 mm diameter tubes (next available size). In the diagram the test measurement with the wider opening is represented by unfilled data points: the initial mass flow rate provided by the controllers has a much lower influence on the remaining parameters of the system. The differential pressure at Pd 1.1 is maximally less than + 10 mbar and at Pd 5.1 less than + 5 mbar. The mass flow rate MFM 5.1 as a result of the initial mass flow controller settings is maximally about 25 mL/min. The spillover diameter of 2 mm is found to be suitable. As equivalent results are expected for Gas Mixing 2, this experiment was not repeated for the second module.

6.2. Evaluation of range for mass flow rate and differential pressure (Pd 1.1 & 5.1)

After the choice of the spillover diameter, it had to be demonstrated in the next step, what range the pumps cover. For a specific setting of gas pump GP 4.1: what is the minimum initial mass flow rate necessary to reach a positive differential pressure at Pd 1.1? And can the resulting pressure increase in the Leaf Chamber be compensated by pump GP 8.1? What is the resulting mass flow rate range? In order to answer these questions, the pump voltage of GP 4.1 was stepwise increased in 0.5 V intervals (0-5 V). Then the following tasks have been performed for every pump voltage setting: First, the mass flow controller set point for CO_2-free air was adjusted manually, so that the differential pressure at Pd 1.1 is approximately + 1 mbar (but clearly higher than 0 mbar). Second, the voltage of gas pump GP 8.1 was adjusted manually, so that Pd 5.1 showed the lowest deviation from 0 mbar. Finally, the mass flow rate was read from MFM 5.1. The results are given in the following diagram (Figure 28), where the blue points are the process values of mass flow controller MFC 1.1, the yellow points are the pump voltages of GP 8.1 and the red points are the resulting mass flow rate measured by MFM 5.1. The mass flow rate is scaled on the primary y-axis; the pump voltage on the secondary axis.

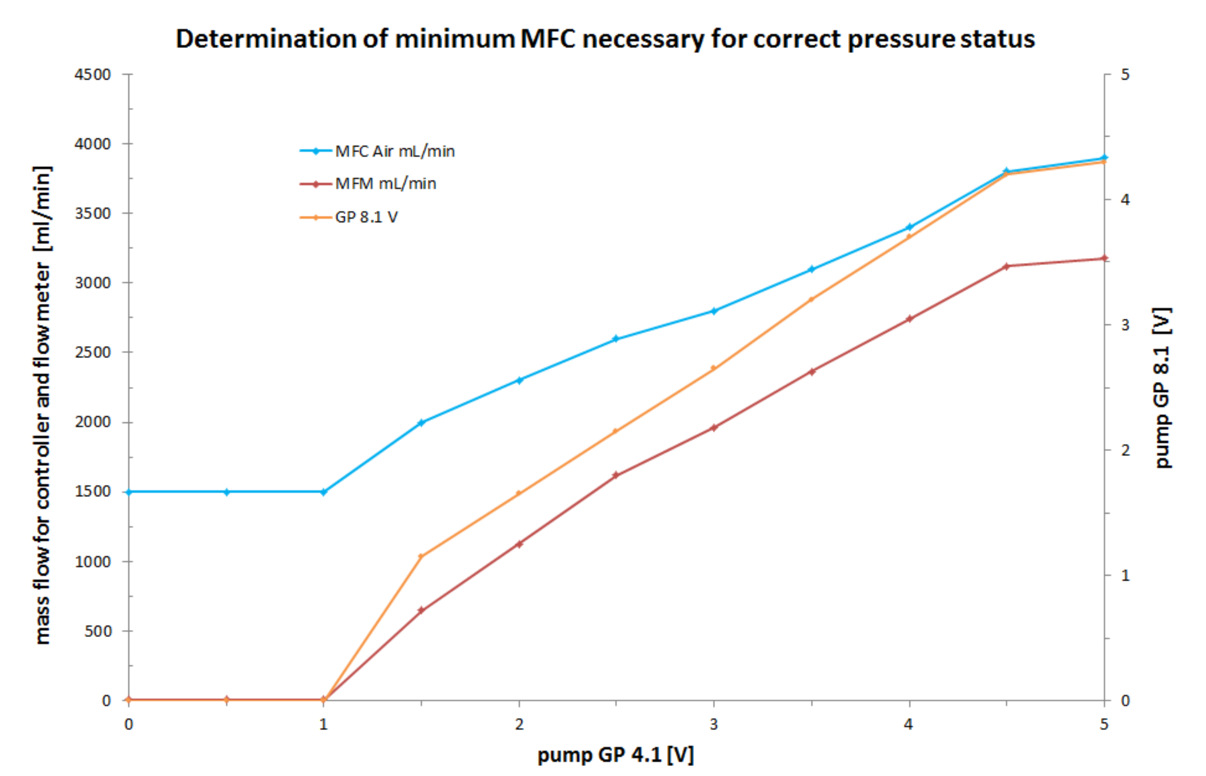


Figure 28: stepwise increase of pump voltage of GP 4.1 and required settings of MFC 1.1 (blue) and GP 8.1 (orange) to maintain the correct pressure status at Pd 1.1 and Pd 5.1

Beginning with lower pump voltages of GP 4.1, it is apparent that the pump has a threshold voltage of about 1 V in order to start pumping. This also has been observed for the other pumps of type NMP085 (bigger pumps). Consequently, the output ranges of the controllers have to be adjusted accordingly. The minimum mass flow rate for MFC 1.1 required to ensure a positive differential pressure at Pd 1.1 was about 1500 ml/min. This corresponds to the amount that is approximately lost through the spillovers. With increasing pump voltage of GP 4.1 the other values increased linearly, as expected. In order to operate at the operating point of 1500 ml/min, a voltage of about 2.5 V for GP 4.1 and 2.0 V for GP 8.1 was required. The mass flow controller set point should be 2500 ml/min at minimum. In the higher voltage region, it is assumed that the maximum power of the pump was reached somewhere between 4.5 V and 5.0 V. The maximum theoretical flow rate for this gas system configuration was 3160 ml/min, which also did not increase when the mass flow controller was set to maximum at 5000 ml/min. However, the gas pump GP 8.1 seemed to have sufficient power to maintain zero differential pressure inside the Leaf Chamber (Pd 5.1). The mass flow controller reference should be at least 4000 ml/min.

However, for this test measurement the influence of GP 9.1 and GP 4.2 has not been considered. GP 9.1 was assumed to have no significant influence on the observed parameters. The diameter of the gas inlet in the Gas Exit and Absorption module will be evaluated at some later point. For GP 4.2 the theoretical maximum flow rate at ambient pressure is 1500 ml/min. In this case, the initial mass flow rate reference for the mass flow controllers must be higher. But for the time being, it should be advised to use small mass flow rates to vent a gas fraction to reference cuvette A of the IRGA.

6.3. Control Loops for mass flow rate and pressure

The difficulty with controlling the mass flow rate and the differential pressure was explained in section 3.1 in detail. Here it should just be recalled that there is positive disturbance of both values on each other so that there is a risk of instability in long terms. In the new setup, three more things have to be considered: First, the pressure increase inside of the system is much higher than in the preliminary system due to the use of significantly more devices. For an operating point of 1500 ml/min a differential pressure of about + 16 mbar at sensor Pd 5.1 was observed with the second controller being disabled. Second, there is a third controller for ensuring a negative differential pressure towards atmosphere at Pd 9.1 by gas pump GP 9.1, which has minor influence on mass flow rate and remaining differential pressure in the system. Third, there are some additional disturbance parameters, which might lead to pressure change, like switching valves and connecting by-pass systems this way. One of these disturbances is venting the gas through the cooling block of the Peltier-element (dew point trap DP 8.1) by switching valves MV 8.1 and MV 8.2. Here, for the operating point an additional pressure increase of + 4.5 mbar was measured.

But before the influence of additional disturbances can be analyzed, the control parameters for the first and second controller had to be evaluated as it was previously done. In the next step, the last controller was added. In this way, the following parameters for a PID type controller were found empirically (Table 5):

	Controller 1	**Controller 2**	**Controller 3**
reference	*MFM 5.1*	*Pd 5.1*	*Pd 9.1*
constant gain K_P	0.001	-0.05	-0.6
integral time T_N [min]	0.05	0.5	1
derivative time T_V [min]	0	0.01	0

Table 5: adjusted control parameters for mass flow and pressure

Usually only the proportional part P and the integral part I are sufficient for the controller. The derivative part D is known for being very sensitive to noise and is rather left out. Anyway, for controller 2 a derivative time T_V was added because it was found that the performance with derivative part is slightly better than without. The following sections will discuss the performance tests done with the control parameters of Table 5. For all test the reference for mass flow controller MFC 1.1 (CO_2-free air or N_2) was set to 4500 ml/min. Valve V 9.1 was kept open. The saving interval was temporarily set to 50 ms to have high time resolution in the diagrams.

6.3.1. System response to change of mass flow rate change

In the first part, the range for the chosen control parameters was tested. Here, the mass flow rate set point has been increased starting from 0 to 3000 ml/min in intervals of 500 ml/min. In consequence, the differential pressure in the leaf chamber increased (measured by sensor Pd 5.1) and was compensated by controller 2 actuating gas pump GP 8.1. The remaining differential pressure sensor values were monitored, too, to prove proper functionality. The results can be seen in Figure 29. Note that differential pressure and mass flow rate are scaled on different y-axis.

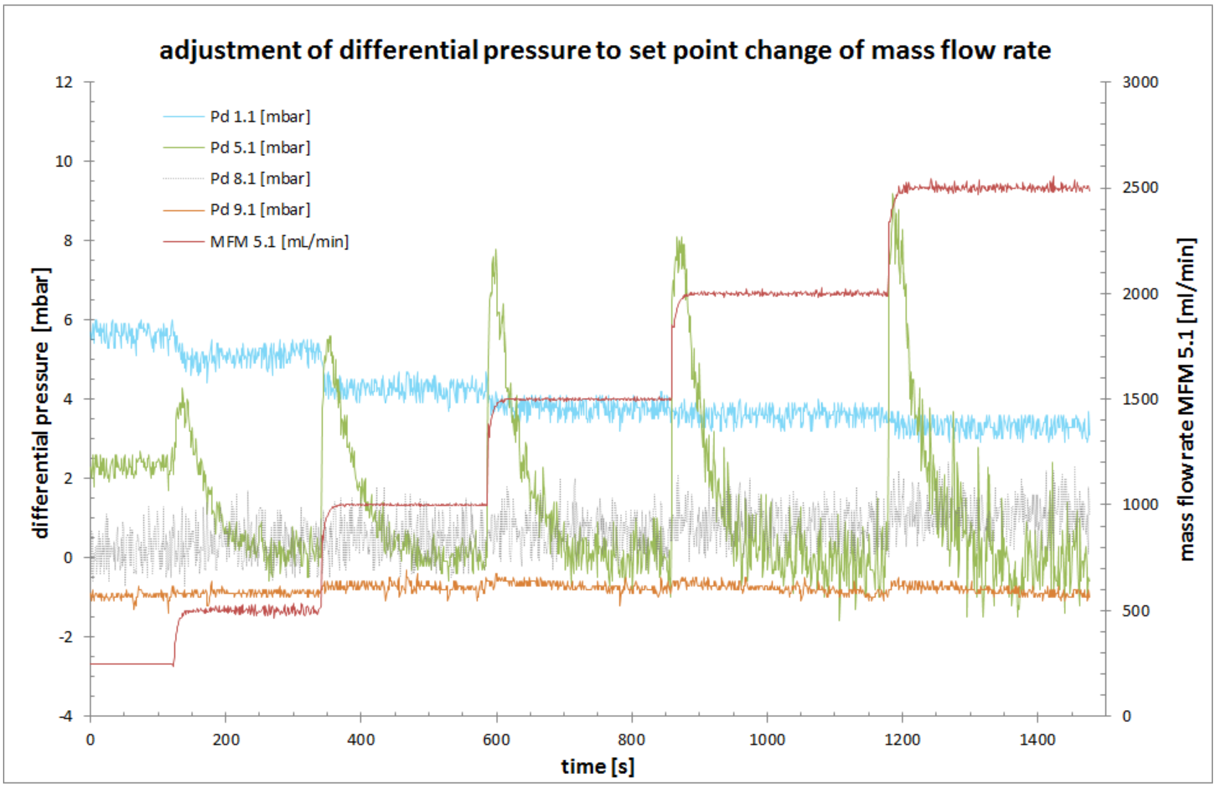


Figure 29: performance test of controller 1, 2 & 3 for mass flow rate set point changes from 500 to 2500 ml/min

The setting controller 1 for the mass flow rate shows good overall performance between 500 and 2500 ml/min, with becoming less good for lower or higher values (fluctuation of ± 50 ml/min). No overshoot or delayed reaction could be observed. In contrast, the corresponding differential pressure course of Pd 5.1 (green line) clearly shows the limitations of the system: the pressure fluctuations after compensation were quite high. This is due to turbulences inside the system, which increase with increasing mass flow rate. They can only be decreased by changing the mechanical setup of the system (e.g. bigger tubing diameter). The fluctuations of the differential pressure Pd 5.1 for the respective mass flow rates are summarized in Table 6, as well as the maximum overshoots. Because of the large fluctuations in Leaf Chamber pressure, it is not recommended to use any mass flows higher than 2500 ml/min for this kind of setup. The performance of the controller 2 is found to be good. Detailed analysis will be conducted for the operating point of 1500 ml/min later. The controller 3 for maintaining a negative differential pressure at Pd 9.1 (orange line) was working reliably and will not be evaluated any further, since there are no significant changes. The differential pressure at Pd 1.1 (blue line) was higher than + 2 mbar throughout the measurement due to the high set point value of 4500 ml/min. The differential pressure at Pd 8.1 was not reaching a critical limit with regard to the LI-7000 and can be neglected.

No performance test was done for decreasing mass flow rate set points, since the same maximum overshoots are expected but in negative direction.

mass flow rate change [ml/min]	max. overshoot at Pd 5.1 [mbar]	fluctuations at Pd 5.1 [mbar]
0 -> 500	+ 4.0	± 0.8
500 -> 1000	+ 5.5	± 0.8
1000 -> 1500	+ 7.5	± 1.0
1500 -> 2000	+ 8.5	± 1.5
2000 -> 2500	+ 9.0	± 2.0

Table 6: max overshoot and fluctuations of differential pressure at Pd 5.1 for increasing mass flow rates

To characterize the response of the controllers to a change in mass flow rate in more detail, the part of the diagram has been extracted from Figure 29, where the reference mass flow has been changed from 1000 ml/min to the operating point of 1500 ml/min (Figure 30). The values for Pd 1.1, 8.1 and 9.1 have been left out for better overview.

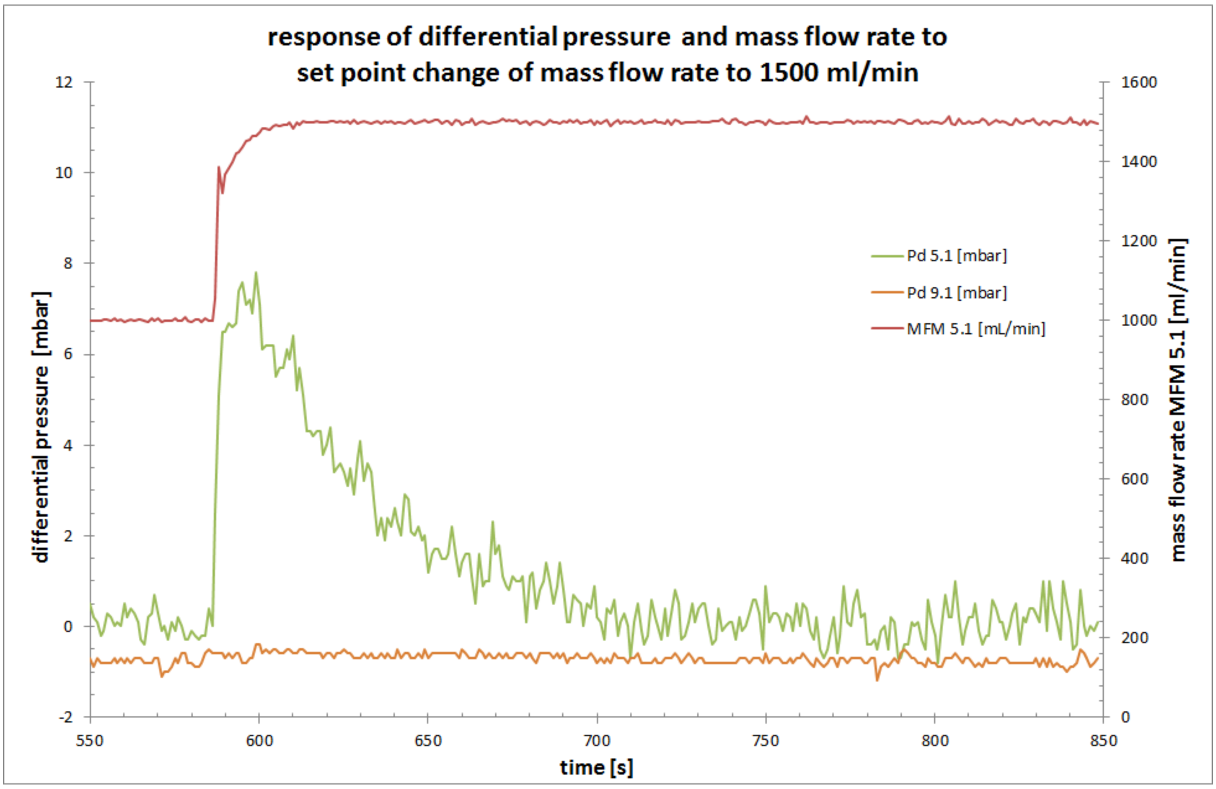


Figure 30: performance test of controllers 1, 2 & 3 for mass flow rate set point change from 1000 to 1500 ml/min (detail)

For the evaluation the following assumption and simplification have been made: The tolerance width for the mass flow rate is ± 100 ml/min and for the differential pressure Pd 5.1 ± 1 mbar (similar to the data point spread). The rise times or recovery times have been estimated including the time to leave the tolerance width for the previous set point (1000 ml/min), since they are much smaller than the respective rise or recovery times. In both cases the rise time equaled the recovery time, since the controlled variable entered the range of the tolerance width and stayed inside. Table 7 summarizes the characteristics of controller 1 for the mass flow rate. Recovery time (= rise time) was determined to be 12 s and no overshoot or permanent offset was observed.

mass flow rate MFM 5.1	
rise time = recovery time	12 s
maximum overshoot	0 ml/min
tolerance width	± 100 ml/min

Table 7: characteristics of performance of controller 1 for the mass flow rate

Table 8 includes the same parameters for the differential pressure at Pd 5.1. The recovery time (=rise time) was about 90 s, not accounting for the fluctuations (average value assumed). The maximum overshoot was about + 7.5 mbar. No permanent offset was observed.

differential pressure Pd 5.1	
rise time = recovery time	90 s
maximum overshoot	+ 7.5 mbar
tolerance width	± 1 mbar

Table 8: characteristics of performance of controller 2 for differential pressure (Pd 5.1)

Since for the mass flow rate a reference change will not be performed during an on-going measurement, settle times and overshoots for both controlled variables are less important than long term offsets. Moreover, a compromise between a high gain for offset compensation and low gain for lower sensitivity against fluctuations had to be found.

6.3.2. System response to a disturbance

As it was mentioned previously, a disturbance can also be introduced by switching MV 8.1 and MV 8.2 at operating point (1500ml/min). The ribs inside the cooling block of the Peltier-element cause additional turbulences and the longer tubing a pressure increase. In order to see the moment the valves are switched, the switching mode of MV 8.1 was also saved to the spreadsheet and displayed in the diagram (purple line, Figure 31). The lower value (0) corresponds to MV 8.1 not being switched and hence by-passing the dew point trap; the higher value (1) indicates the detour through the dew point trap. The diagram shows the time course of mass flow rate measured by MFM 5.1 and differential pressure at all relevant pressure sensors. As can be seen from the graph, the detour does not affect the mass flow rate (red line). The pressure inside the Leaf Chamber (Pd 5.1) reacted with a slight increase if passing the cooling block and with a slight decrease if it was by-passed again. The fluctuations became more severe due to more turbulence. Assuming an average, the maximum overshoot is about ± 1 mbar. No permanent offset was observed. Since the tolerance width was defined to be ± 1 mbar, it was impossible to determine a rise or recovery time due to the differential pressure staying within the defined width on average. However, it can be said that after about 60 s the pressure normalized to ambient pressure again. The reactions of all other sensors were not significant and can be ignored. The difference of the pressure at Pd 9.1 towards zero was found to be very small, so the reference for Pd 9.1 is changed to -5 mbar and the constant gain of the proportional part was set slightly higher (K_P = -0.8). In summary, the parameters for the controllers are found to be suitable.

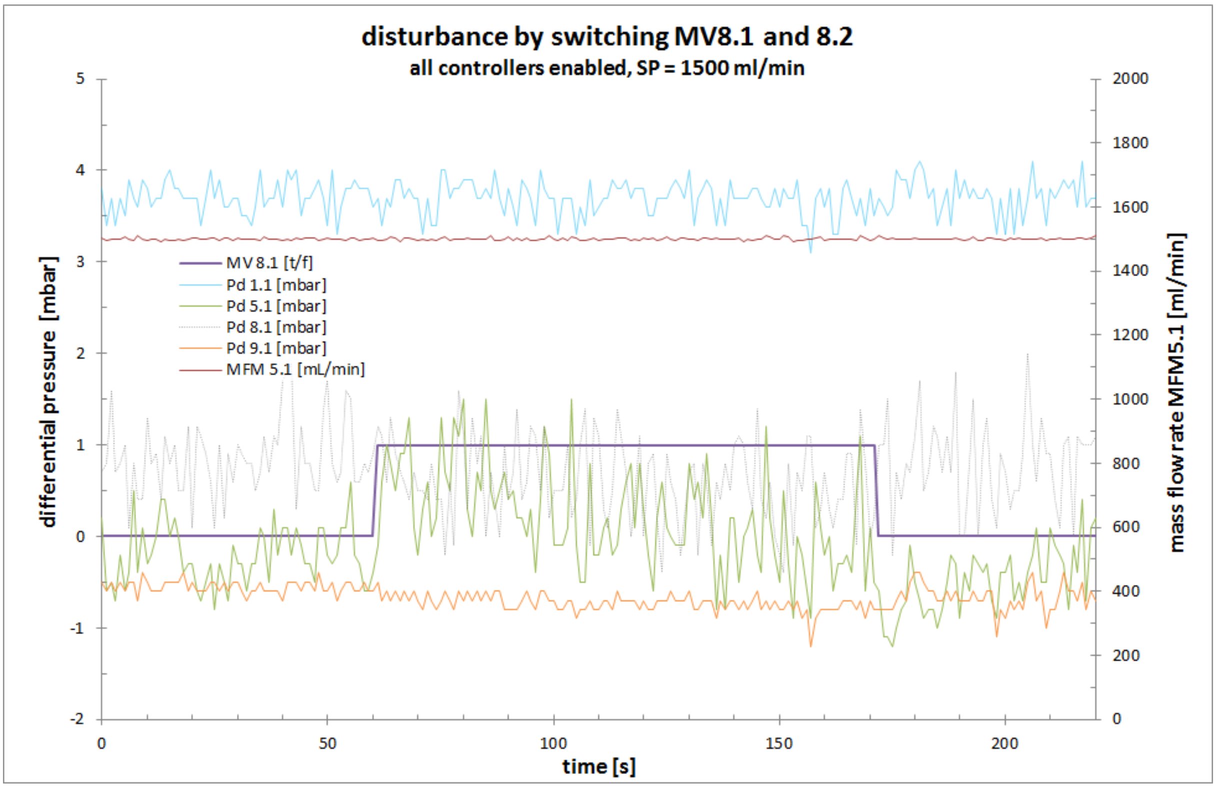


Figure 31: response of controlled variables to disturbance introduced by passing the dew point trap DP 8.1

6.4. Adjustment range for CO_2 mass flow rate

For future studies on gas exchange of plants a wide range of CO_2 concentrations should be covered, allowing also studies under elevated atmospheric CO_2 concentrations to estimate effects of global climate changes on plants. Currently the atmospheric CO_2 concentration is about 400 ppm (Dlugokencky, 2013) or 0.04%. This concentration can e.g. be reached by setting the reference of the mass flow controller for CO_2 free air (MFC 1.1) to 4000 ml/min and the reference of the mass flow controller for CO_2 to 1.6 ml/min. At a fixed mass flow rate for N_2 at 4000 ml/min a test has to be done to find about the possible range for CO_2 concentration. It is assumed that the inject nozzle (Bronkhorst-Mättig, Germany) is designed for specific flow rates of CO_2 and limit the concentration range. Furthermore it is to be determined to what extend the adjusted CO_2 concentration can be mirrored with the IRGA.

Before the measurement was performed, the reference cell A of the IRGA (LI-7000) was calibrated with dry N_2 gas (manual mixing mode). Then the measurement cell B was calibrated to match reference cell A. For this purpose multivalve MV 8.3 had to be switched. The humidification part of the Gas Mixing module 1 was bridged for calibration and measurement by switching MV 1.1 and 1.2.

For the test measurement the reference of the mass flow controller MFC 1.1 for N_2 was kept constant at 4000 ml/min (MFC 1.2 for O_2 was set to zero), whereas the reference of MFC 1.3 for CO_2 was increased stepwise by 0.5 ml/min. The reference mass flow rate was set to the operating point of 1500 ml/min. The small gas pump GP 4.2 pumped maximally at about 1500 ml/min (12 V), so that the reference gas fraction reached cell A with least delay, maximizing the delay of the remaining gas fraction for measurement cell B. In this way the maximum delay between reference and measurement gas stream, caused by the longer way to cell B, can be determined.

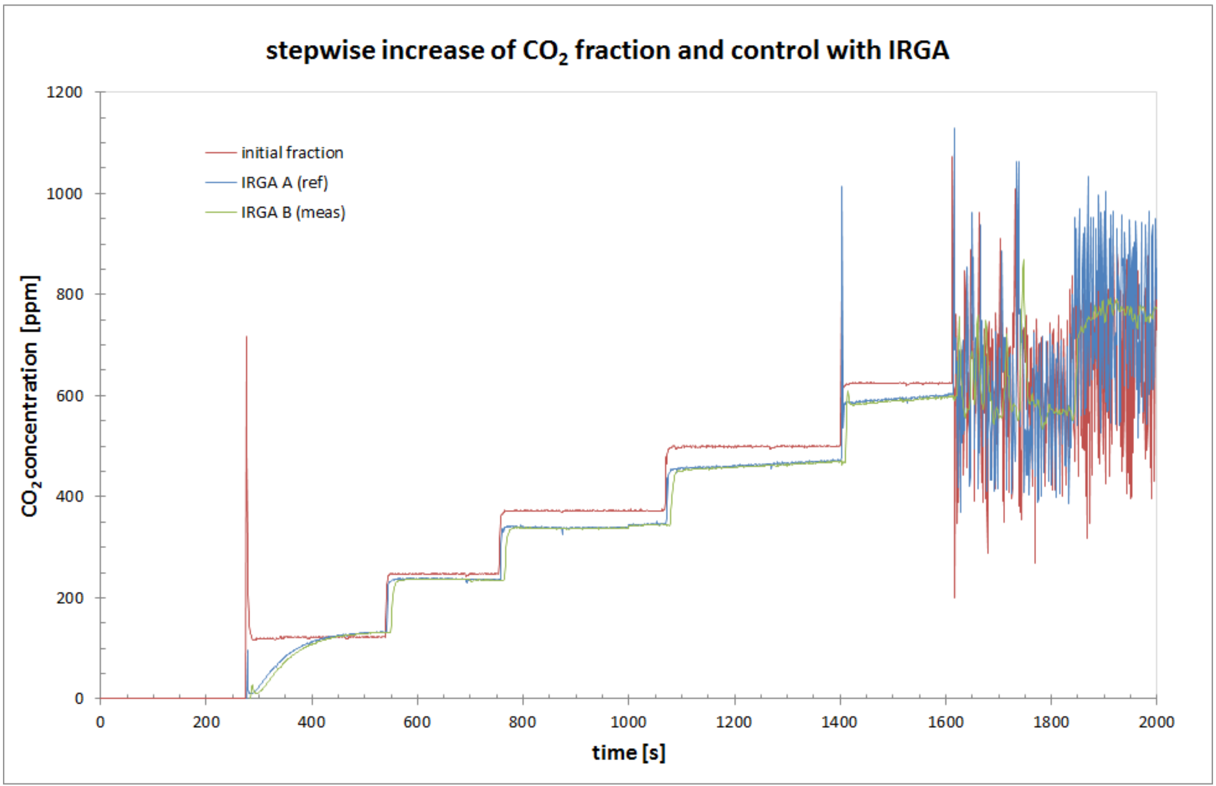


Figure 32: stepwise increase of initial CO_2 fraction and monitoring the changes with the IRGA

Figure 32 shows the time course of the initial fraction of CO_2 (red line), the CO_2 concentration of reference cell A (blue line) and measurement cell B (green line) of the IRGA in units of ppm. At a flow rate of 0.5 ml/min (corresponding to 125 ppm), the mass flow controller reacted with a strong overshoot to the set point change and adjusted quickly thereafter. It took about 150 s until the measurement values of the IRGA had reached their maximum at this set point. This might be due to dead volumes and adsorption of CO_2 once inside the system. For higher flow rates of CO_2 no such delay could be observed. With increasing CO_2 concentration the deviation between initial and measured CO_2 fraction increased. This can have several reasons like adsorption of CO_2 on system components, interaction with dead volumes or missing 2-point calibration. But apart from that no significant offset between cell A and B of the IRGA could be noticed in the steady state: the difference spread between 2 and 4 ppm. A proper calibration will allow for a precise detection of CO_2 uptake/emission of a plant leaf (delta measurement). On average, it took about 2 s until a change in initial CO_2 fraction could be detected in reference cell A, and about 13 s for measurement cell B. This means that the longer way through the Leaf Chamber caused a measurement delay of about 10 s. For flow rates higher than 2.0 ml/min (corresponds to 500 ppm) the mass flow controller could not adjust to the correct reference value anymore and reacted with very large fluctuations (measurement was stopped at 3.5 ml/min of CO_2). This is probably due to increasing turbulence at the inject nozzle of the gas mixer provided by the manufacturer of the MFCs (Bronkhorst-Mättig, Germany), which combines the gas streams from both mass flow controllers, but there might be other reasons as well (gas supply). To increase the fraction of CO_2 it is therefore suggested to temporarily reduce the set point of the mass flow controller for CO_2-free air/N_2 or O_2 as much as possible, so that a positive differential pressure is still guaranteed at Pd 1.1 or 2.1.

6.5. Determination of CO_2 dwell time

For calibration purposes it might be interesting to know the dwell time of CO_2 inside the system once the gas supply has been changed to N_2 or CO_2-free air only. This time has to pass at minimum before the IRGA can be calibrated against zero CO_2. To determine the dwell time, the MFC 1.2 for CO_2 was set to its maximum at 5.0 ml/min and the MFC 1.1 for N_2 to 4000 ml/min: This corresponds approximately to a CO_2 fraction of 1250 ppm. The mass flow rate MFM 5.1 was set to the operating point of 1500 ml/min throughout the entire measurement. The humidification part of the Gas Mixing module was by-passed again. V 9.1 was kept open as usual and all other valves were set to default. The time course of the CO_2 decrease is presented in Figure 33. The red line represents the initial fraction of CO_2, which has been changed to zero after about 70 s. The measured values of the IRGA are given in blue (cell A) and green (cell B).

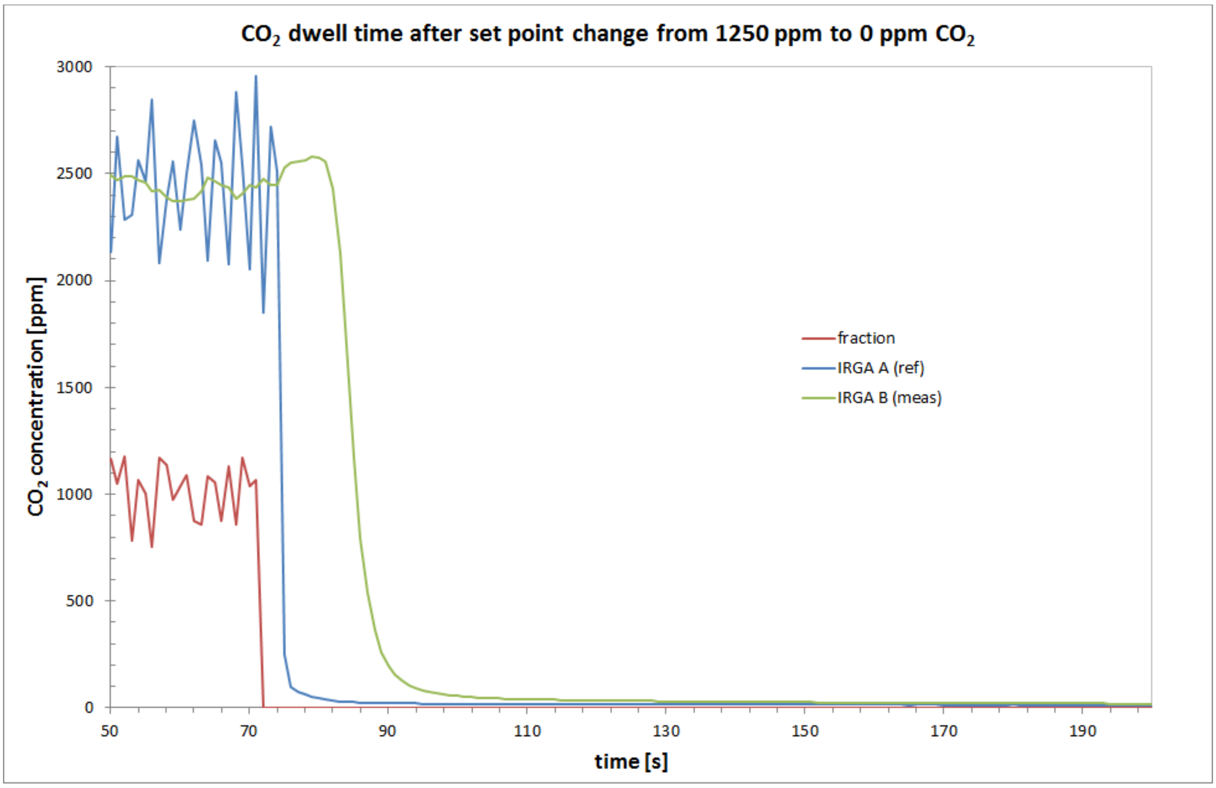


Figure 33: time course of CO_2 concentration after set point chance from 5.0 ml/min (1250 ppm) to 0 ml/min at initially 4000 ml/min of N_2 and at operating point (1500 ml/min)

First of all, it strikes that the CO_2 concentration measured by the IRGA (cell A and B: about 2500 ppm) was about double the amount, which is calculated from the mass flow controller values for CO_2 (about 4 ml/min) and N_2 (4000 ml/min): 1000 ppm. Reasons can currently not be given and further evaluation is required for higher concentrations of CO_2. However, one can be sure that a maximum concentration of CO_2 is given at this operating point, making the measurement even more significant. The CO_2 fraction of cell B was not subject to the same fluctuations of cell A due to buffering effect of the longer gas way through the Leaf Chamber. After about 70 s the MFC set point for CO_2 was changed to zero promptly. The initial fraction of CO_2 (red line) quickly dropped to zero. The CO_2 concentration in cell A of the IRGA quickly decreased accordingly after about 2 s (blue line), but did not go down to 0 ppm. The concentration in cell B reacted similar with a delay of about 15 s (green line). CO_2 remained inside the system for quite a long time, decreasing only very slowly. The concentration of both cells never dropped below 1 ppm during 17 min (entire measurement not shown). Most probably the gas was adsorbed on surfaces of system components or stuck in dead volumes. Consequently for calibration purposes, the gas system should be vented with N_2 for at least 20 min. Because the dwell time of the gas also depends on the chosen mass flow rate, it is advised to extend the venting period.

6.6. Adjustment range for humidification

In order to simulate different environmental conditions, it is also required that the humidification can be adjusted in a broad range. This is provided by the dew point traps inside the water bath of the refrigerating circulator. Initial tests were already performed (chapter 3.2 & 3.3). So here, just the minimum and the maximum humidity should be determined. Furthermore the correlation with humidity sensing apparatuses of the Main Gas Cycle (IRGA, humidity sensors H 5.1 and H 5.2) can be observed.

For the test run, dry N_2 was adjusted to a mass flow rate of 4500 ml/min (manual mixing mode). The mass flow rate was set to 1500 ml/min (operating point). The small gas pump GP 4.2 was operated on 6 V. The heater mat was powered and the tubing was connected to the peristaltic pump. Three set points were chosen for the water bath temperature: the first one yielding maximum humidification (30°C = temperature of the surrounding environment); the last one yielding minimum humidification (1°C = coldest temperature for water bath) and an intermediate humidity (16°C). The measurement was conducted starting with the higher temperature and decreasing it stepwise. For each temperature set point time was given to adjust to steady state conditions. The time course of the measurement can be seen in Figure 34. Relative humidity and dew point temperature have been scaled on the primary y-axis, whereas the temperature of the humidity sensors is scaled on the secondary y-axis. In this way it was avoided that the curves do not lie on top of each other.

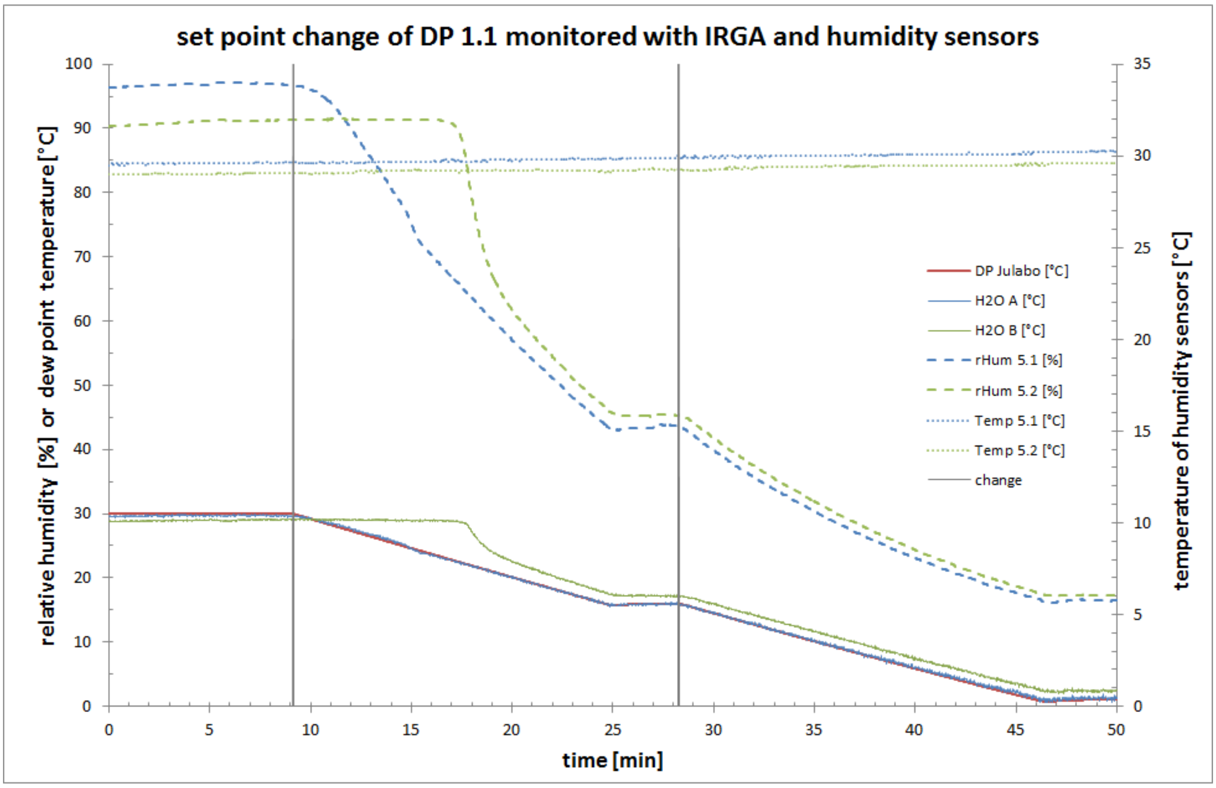


Figure 34: change of initial dew point temperature & monitoring of humidity in the Main Gas Cycle (IRGA, H 5.1 & 5.2)

Each moment of set point change is indicated by a grey vertical line. The bath temperature is given by a red line. All parameters upstream of the Leaf Chamber are presented in blue, all downstream of it in green. The dotted lines are values obtained from the humidity sensors (H 5.1 and H 5.2). The humidity values measured by the IRGA are given in units of dew point temperature (°C) for easier comparison and are shown in continuous lines.

At a water bath temperature of 30°C, the humidity sensors as well as the IRGA measured a lower humidity on the output side (90 %) than on the input side (96 %). Looking at the temperatures, it is obvious that the gas slightly cooled down by passing the Leaf Chamber. Within the 19 inch rack, a lot of heat is emitted by several devices. The Leaf Chamber is positioned in the front of rack, where it is subject to slightly cooler temperatures and draft. This suggests that due to the lower temperature water vapor condensed inside the Leaf Chamber. (During an additional longer run, the glass of the Leaf Chamber steamed up after a while, proving the above assumption.) After changing the set point of the water bath to 16°C, the water bath adjusted slowly. The humidity before the Leaf Chamber reacted accordingly; the lines of the dew point of the water bath and cell A of the IRGA are even congruent. At the beginning of the change the humidity on the output side did not change, building a hump in the curve. This is due to the condensed water, which has to evaporate, first of all. This is completed after about 10 min; from then on the lines of the input and output side go in parallel. The new dew point set point (16°C) was reached after about 16 min, indicated by the second plateau. The corresponding relative humidity was about 45 %. When the set point was changed to 1°C, the water bath temperature adjusted within 18 min. The corresponding relative humidity was about 18%. Concluding, the humidity range is about 18% to 90% relative humidity.

6.7. Evaluation of dew point trap DP 8.1

The dew point trap DP 8.1 is enabled to eliminate water vapor transpired by a leaf inside the Leaf Chamber. The difference in humidity, which is to be eliminated, is detected by the IRGA (cell A and B). The initial tests described in chapter 3.3 were performed under different conditions than this is the case now: Temperature controllers are enabled instead of the Power Supplies and the heater mat unit has been removed. This is suspected to influence the process, so a step response from 0 % to 100 % and back to 0 % is performed again, to characterize it. According to the characteristics of the process, the parameters for the LabVIEW controller will be defined and tested. In the course of this, it is to be determined, what cooling range in terms of dew point temperature of the gas the Peltier-element covers and how precise humidity can be eliminated, which emerges in the Leaf Chamber.

6.7.1. Choice of controller parameters

For acquiring a step response, CO_2-free air streamed with 1500 ml/min through the system. The small gas pump GP 4.2 ran at 6 V. The valves MV 8.1 and MV 8.2 were enabled in order to vent gas through the dew point trap DP 8.1. The remaining valves were set to default (with valve V 9.1 being open). The step response was not recorded with LabVIEW, but with software provided by manufacturer in parallel, because this part was not yet implemented into the LabVIEW program. As feedback signal, the temperature of the cooling block measured by the NTC is taken. Later, the program was modified in such a way that the dew point temperature of the gas was controlled by LabVIEW using the dew point of cell B of the IRGA. However, a similar reaction of both feedback signals is assumed (a little delay can be ignored), so that a step response measurement was not repeated after the modification of the LabVIEW program.

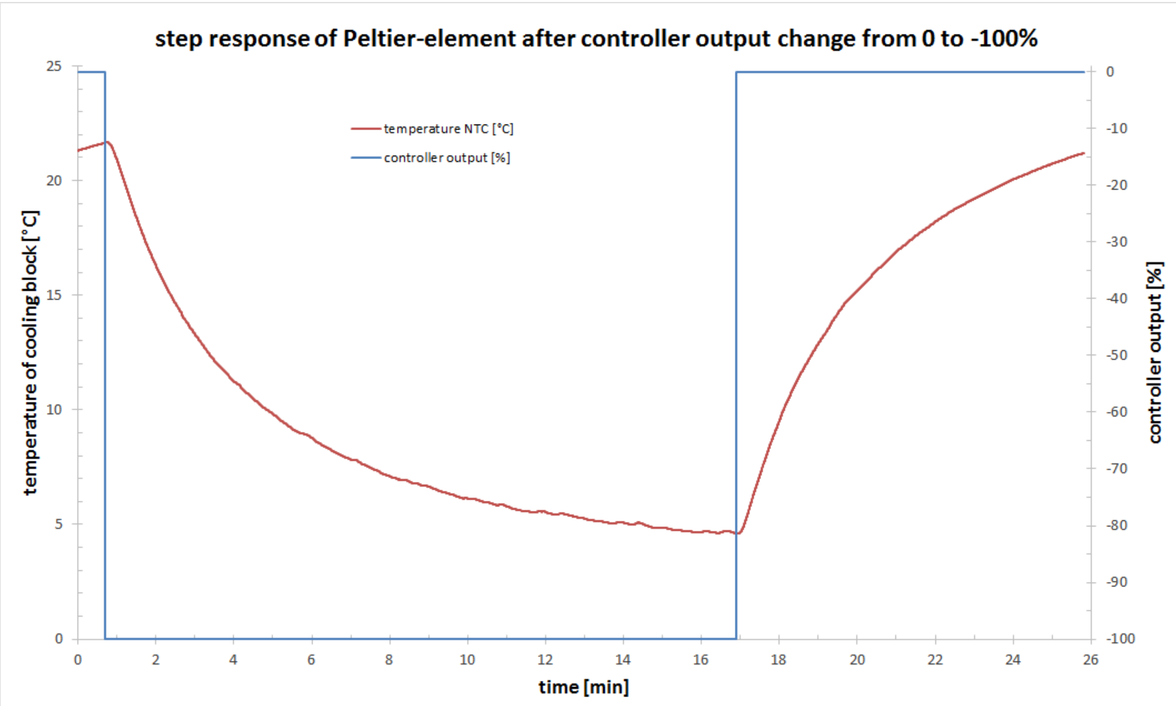


Figure 35: temperature course of cooling block (DP 8.1) after controller output change from 0 to -100%

Note that maximum cooling corresponds to -100% for the temperature controller. The step response is given in Figure 35. The controller output is presented in blue (scaled on the secondary y-axis); the temperature of the NTC in red (scaled on the left y-axis). First, the controller was set to maximum cooling. Afterwards the Peltier-element was allowed to warm up. By drawing a tangent in the inflection point, equivalent dead time T_u and settling time T_g were determined for the cooling phase and warm-up phase. The proportional gain of the process was estimated to be 1. The proportional gain of the controller K_P* was calculated based on the reciprocal of the proportional gain of the process (= 1) and weighted by the ratio T_g/T_u. The results are listed in Table 9 and Table 10 on the left side.

cooling:

characteristics of process		parameter for PID controller	
K_P*	20	K_p	19
T_u	0.2 min	T_N	0.5 min
T_g	4 min	T_V	0.08 min

Table 9: process variables and new parameters for a PID controller in the cooling phase

warming up:

characteristics of process		parameter for PID controller	
K_P*	30	K_p	29
T_u	0.1 min	T_N	0.2 min
T_g	3 min	T_V	0.04 min

Table 10: process variables and new parameters for a PID controller in the warm-up phase

The parameters for a PID controller were determined according to the tuning rules of Chien, Hrones and Reswick for aperiodic behavior and disturbance response (Lutz & Wendt, 2010). They are listed on the right side of the corresponding tables (Table 9, Table 10). The performance test of the chosen controller parameters is discussed in the following section 6.7.2.

In a separate test, the controller was set again to maximum cooling (-100 %) to determine the minimum temperature of the cooling block. In the steady state, the NTC monitored a temperature of 4°C. The corresponding dew point temperature of the gas is assumed to be a little higher due to efficiency loss.

6.7.2. Performance tests

The amount of water evaporated by plants varies a lot and is highly regulated with respect to internal and external conditions of the plants. In order to increase the humidity in the Leaf Chamber in a defined way, water was injected into the Leaf Chamber by a simple device. It consisted of a piece of tissue and a Teflon capillary, which was lead into the Leaf Chamber by a third opening (Figure 36). The capillary was connected to a needle and a syringe filled with water. The space between capillary and tube was sealed additionally.

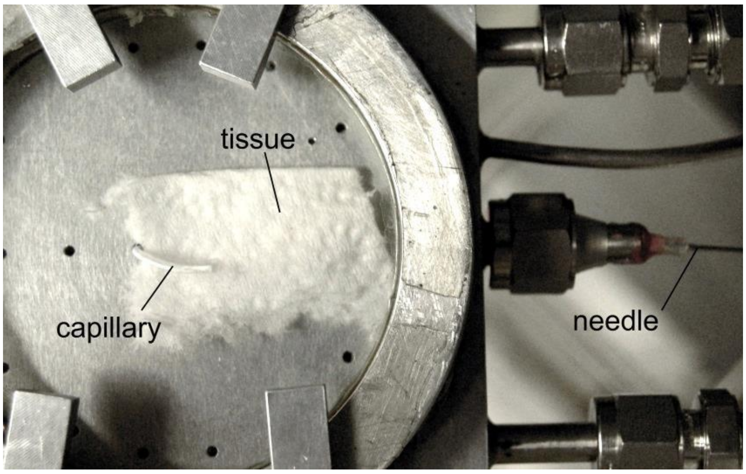


Figure 36: administration of water for addition of artificial humidity in the Leaf Chamber

By dripping in water, the humidity inside the Leaf Chamber can be increased significantly at once. This is probably much more than a single plant leaf transpires, but it is a good method to imitate a worst case scenario.

The performance of the controller with the chosen parameters was tested after the changes have been made in the LabVIEW program (direct control by LabVIEW). All parameters in the program were set as before at operating point. But the water transport cycle for the removal of water inside of the cooling block (DP 8.1) had to be modified, since too much water was trapped inside the gas line and had to be eliminated. Instead of the closed water cycle, air from the water reservoir was blown in and the condensed water was sucked out again. Because this bears the risk of wrong measurement and, potentially, radioactive contamination by exchange with air of the Gas Mixing module, it has to be replaced with the old cycle (Figure 14) or a new water removal system afterwards.

The reference dew point temperature at the refrigerating circulator (DP 1.1) was adjusted to 23°C initially, and to 16°C and 8°C successively. The Peltier-element was enabled before the saving of the data was started to allow for settlement. Water was dripped into the Leaf Chamber before and during the data logging.

The set point of the water bath and the resulting reaction of all devices measuring humidity are displayed in Figure 37. The temperature of the humidity sensors H 5.1 and 5.2, of the cooling block (measured by the NTC), and of the water bath, as well as the dew point temperatures of the gas measured by the IRGA (cell A and B) are scaled on the primary y-axis. The relative humidity is scaled on the secondary y-axis. The moments, where the set point of the refrigerating circulator has been decreased, are indicated with grey vertical lines. The temperature of the water bath itself is plotted in red; the one of the cooling block in purple. Again blue lines are chosen for parameters acquired from devices upstream of the Leaf Chamber and green lines for those downstream of the Leaf Chamber. Dotted lines refer to the humidity sensors H 5.1 and 5.2. The dew point temperatures in cell A and cell B of the IRGA are presented with continuous lines.

As can be seen from the relative humidity of H 5.1 and 5.2, the relative humidity downstream of the Leaf Chamber was higher than before. After about 27 min a sudden increase in humidity (H 5.2) indicates the injection of water into the Leaf Chamber. The temperatures of H 5.1 and 5.2 were constant but differed in about 1°C.

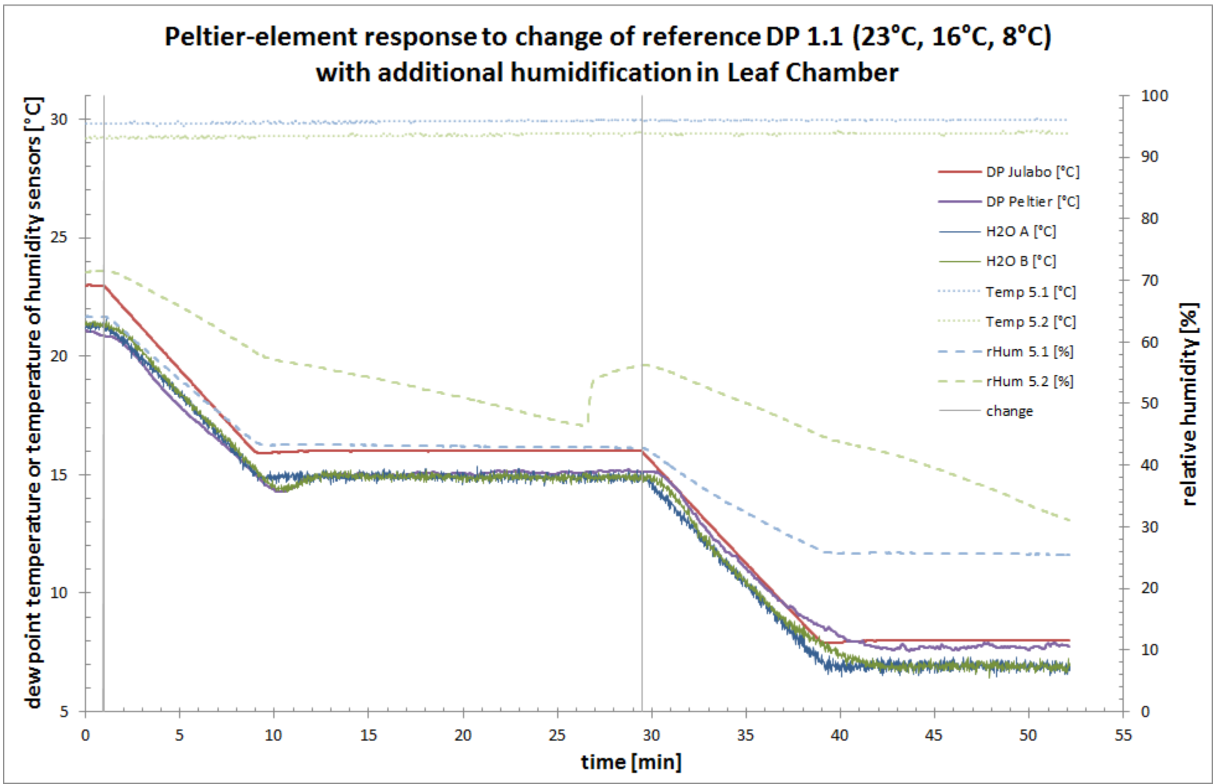


Figure 37: Influence of set point change of water bath (DP 1.1) & addition of humidity in the Leaf Chamber on DP 8.1

The dew point temperature of the gas measured by cell A of the IRGA was about 1°C to 2°C lower than the water bath temperature of the refrigerating circulator probably due to the increase in temperature of the gas. The dew point temperature measured by cell B followed this course with two exceptions: there's a slight overshoot of about 1°C, when the temperature of the water bath adjusts to 16°C (at about 10 min). And when the refrigerating circulator has adjusted to the lower temperature of 8°C, the dew point temperature measured in cell B approached the target temperature with a longer settling time. But since no set point changes are expected to be made during a measurement with a plant in future, deviations resulting from a set point change can be ignored. Here, first of all the influence of a disturbance input is important. When the humidity was increased artificially after about 27 min, neither an initial increase of the dew point temperature in cell B nor an influence on the temperature of the cooling block could be observed. It is assumed that the efficiency factor of the energy transfer from the gas to cooling block is very good and therefore high enough to condense any artificially added water vapor of the gas. For this reason, it might be better to optimize the controller parameters for command response (set point change of the DP 1.1). But for the time being the choice of the controller parameters serves its purpose, since no steady state offset is observed and the control loop behaves stable.

Other disturbances can be e.g. the change of the mass flow rate, which has a slight influence on the equivalent dead time or settling time of the influence. A test with a step from 1500 ml/min to 2500 ml/min did not yield any significant change. A decrease in humidity in the Leaf Chamber is not expected during normal measurements and hence will not be considered.

7. Summary

In plant research it becomes increasingly important to quantify plant growth in the context of phenotyping. In order to study physiological mechanisms like the uptake of carbon dioxide and conversion into organic molecules, non-invasive methods need to be applied. One of these methods is to administer radioactively labeled CO_2 to plant leaves and to monitor the uptake and distribution of radio tracers inside of the plant. For this purpose, a special multifunctional system has been set up to monitor the gas exchange of an intact plant leaf. The system delivers a defined gas composition variable in gas concentrations of CO_2-free air (or a mixture of N_2 and O_2), CO_2 and humidity to a plant leaf enclosed in a cuvette. The uptake of CO_2 and the released water vapor caused by the leaf's activities (photosynthesis and transpiration) can be monitored with an Infrared Gas Analyzer. The differential pressure inside of the cuvette and mass flow rate in the gas exchange system are subject to control. On demand, water vapor transpired by the plant can be eliminated by a dew point trap before the Analyzer. The gas exchange system can be operated as open system (continuous supply of fresh gas and disposal) and as closed system (leaf disconnected from fresh gas supply and disposal). In the closed mode, the system should allow the administration of radioactively labeled $^{11}CO_2$ (positron emitter with a half-life of 20 min) to a plant leaf. Since the radioactive application part will go beyond the scope of this thesis, it has not been implemented here, but was considered in the planning.

The setup of the gas exchange system included the mechanical and electrical assembly of all system parts in a 19-inch rack and the software implementation with LabVIEW. It has been taken care that space-saving solutions have been applied, system parts are aligned according to their technical requirements, gas lines are as short as possible, smallest dead volumes are present, inert materials are in use, and radioactive safety is guaranteed. The LabVIEW program interface provides a comprehensive overview and intuitive handling of the system. In order to approve the functionality and to identify the limitations, evaluation tests have been made. Currently, the following settings are possible:

- The mass flow rate can be adjusted in a range of 0 to 2500 ml/min, allowing measurements with small leaf chambers (≈ 40 ml). Mainly, the mass flow rate is limited by the differential pressure increase and turbulences due to the small inner diameter of the recent tubing (4 mm).
- The CO_2 concentration can be varied between 0 and 600 ppm. This is much less than expected, but can be optimized by the replacement of components.
- The initial humidity in terms of dew point temperature of the gas can be set between 1°C and room temperature (corresponding to about 15 to 90 % relative humidity) and is found to cover a sufficient range. However, if humidity transpired by a plant leaf needs to be eliminated, the dew point temperature can minimally reach 4°C (about 20 % relative humidity).

During the operation of the gas exchange system, some problems have been noticed: The space is still very limited, so that the $^{11}CO_2$-application module can currently not be implemented *inside* of the rack. Most of the devices inside of the rack release a lot of heat, which also affects the gas lines. The Gas Generator is defective and needs to be replaced. The dwell time of CO_2 inside the gas lines is about 30 min. For potential users it should be noted, that some devices still require manual operation (switching MV 4.1 adjustment of gas pump GP 4.2, powering of heater mats, alignment of water tubing). In future, it is planned to implement a pulse width modulation unit to actuate the small gas pumps based on LabVIEW, too. Further, the system is to be expanded by installing the $^{11}CO_2$-application module. Then the next steps will be to administer some $^{11}CO_2$ as part of a leakage test as well as to evaluate the functionality of the whole system with an enclosed plant leaf.

With the multifunctional system constructed in this project, it can be studied how different environmental conditions influence plants. It is possible to apply a defined gas composition and to observe the exchange with a plant leaf. It is also dedicated for the application of radioactively labeled CO_2 and therewith will enable tracer studies in the course of phenotyping.

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10. List of abbreviations

Abs	Absorber
DP	Dew point trap
FI	Filter
GP	Gas pump
H	Humidity sensor
Hm	Humidifier
IRGA	Infrared Gas Analyzer
K_P	Proportional gain of controller (as part of controller parameters)
K_P*	Proportional gain of controller based on proportional gain of the process
LC	Leaf Chamber
MC	Motor controller
MFC	Mass flow controller
MFM	Mass flow meter
MV	Multivalve
NV	Needle valve
PA	Ambient pressure sensor
Pd	Differential pressure sensor
PRP	Pressure reference point
QC	Quick connectors
RD	Radio detector
SP	Set point
TC	Temperature controller
T_g	Settling time
Th	Thermostat
T_N	Integral time
T_u	Equivalent dead time
T_V	Derivative time
V	Valve
VM	Valve motor

---Attachments have been deleted---