

SEQUENTIAL FLUID INJECTION PACK

DOCUMENT REF: UG121-0715

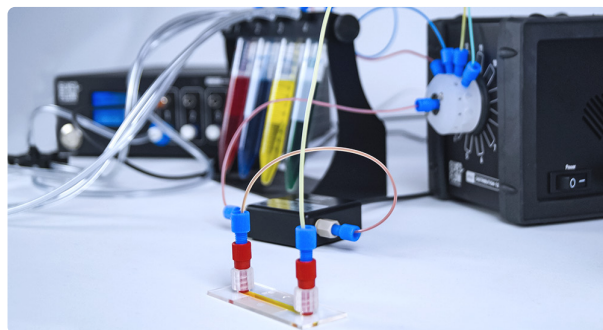
Introduction	2
Content of the sequential injection pack	3
Before starting the experiment	7
Let's start injecting different solutions sequentially!	10
Troubleshooting	15
Going further - Appendices	16
Microfluidic connector guide	16
Flow rate control	19
Volume control	23
System automation	25
Serialisation/scale up - working with more than 12 solutions	28
Stop the flow in the device	30
Optimization of your sequential injection system using valves	31
Work with tiny volumes	34
Lower the impact of the mechanical switch	35
Automated control of the injected volume using the "flow integration" module	36
Supplementary information	38

Introduction

The **sequential injection pack** offered by Elveflow is designed to allow optimal use by anyone, whether they are already familiar with microfluidics and fluid handling automation or have never used it before. Simple and intuitive instructions are provided to quickly and easily inject various solutions or samples sequentially in a microfluidic line and control the injected volumes.

When starting out, the user can follow the provided protocol step by step to **inject sequentially up to 12 solutions in a device**.

Numerous tips are provided throughout the document. Finally, the appendices provide numerous information to master the injection technique, automate the process and control the injected volumes to fully master the versatility offered by this sequential injection system.



The sequential injection system involves two principal elements: a **pressure controller** and a **rotary valve**. The function of the pressure controller is to accurately pressurize the different reservoirs containing the various liquids. Once pressurized, the liquids can be connected to the rotary valve which is used as a selector. The rotation of the valve allows the user to choose the liquid which goes through the valve into the system. Refer to the following videos to visualize the [working principle](#) and [outcome of such injection](#).



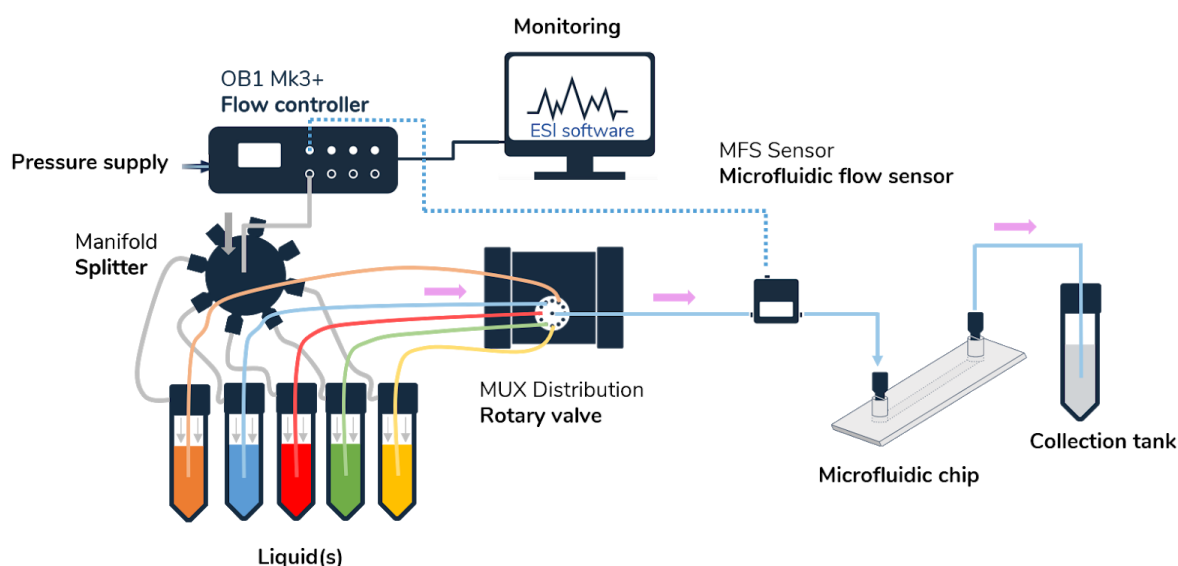
Four different aqueous solutions spaced by oil

The setup presented in this user guide is an example where **5 different solutions** are injected in a microfluidic chip. This flexible and versatile setup can be adapted to any user requirement. Tips and tricks are given throughout the document to tailor the setup and for example adapt the number of solutions injected, automate the system and set up a fast liquid swap, etc.

If your requirements are not fully met using the information provided in the guide, please feel free to contact us at contact@elveflow.com.

Non exhaustive list of applications for which the sequential injection pack can be used:

- > Cell culture under flow
- > Functionalization, characterization or test of sensors
- > Calibration of sensors
- > Toxicity or drug screening
- > Gradient assay
- > Perfusion
- > Fluorescence In Situ Hybridization (FISH)
- > Any application that involves automated and/or fast medium switching



Schematic of the sequential injection setup for the injection of 5 different solutions in a microfluidic chip

Contents of the sequential injection pack

1 OB1 MK3+ Pressure controller

1 channel with 0 to 2 bar range

The pressure controller is the centerpiece of the setup; it enables the precise control of the pressure difference across the fluidic system and thus accurate control of the liquid flow.



Other pressure ranges are available, also including vacuum control. Depending on the requirements of your experiment, you can choose between different pressure ranges : 0 to 200 mbar, 0 to 2000 mbar, 0 to 8000 mbar, -900 to 1000 mbar and -900 to 6000 mbar.



PRINCIPLE OF PRESSURE-DRIVEN FLOW RATE CONTROL

Elveflow instruments use pressure to drive liquid flows in fluidic systems

The elementary principle is that a pressure difference across a fluidic line (between the outlet and the inlet of the fluidic system : $\Delta P = P_{out} - P_{in}$) generates movement of the liquid in the system characterized by its volumetric flow rate. The flow rate Q is proportional to the pressure difference, and the proportionality coefficient R is called the fluidic resistance:

$$\Delta P = R \times Q$$

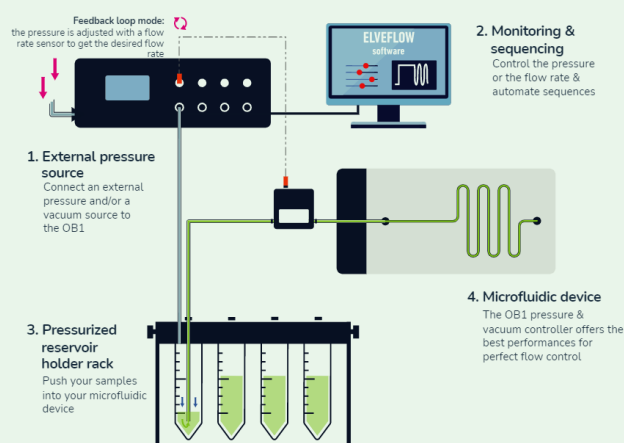
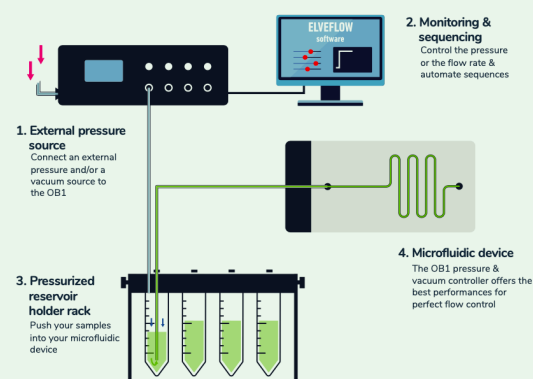
R depends on the geometrical characteristics of the fluidic system and on the properties of the liquid.

For instance: the longer or the thinner the tubing or the more viscous the liquid, the greater the resistance. It means that a higher pressure difference is required to obtain the same flow rate in a system with longer or thinner tubing or with a more viscous liquid.

For instance the resistance of a circular cross channel is $R \propto \frac{\mu L}{r^4}$ radius

Each element of the circuit (chip, tubing, sensors, ...) contributes to the whole resistance R of the system.

The **OB1 pressure controller** creates and controls the pressure difference by pressurizing the air in a sealed reservoir that contains the liquid at the inlet of the system. The outlet pressure Pout most often equals the atmospheric pressure, so the OB1 controls ΔP .



A **flow rate sensor** can be added in line to measure and monitor the flow rate Q .

IN PRESSURE CONTROL: the user controls the pressure difference (the command is a pressure) and it generates a flow rate.

IN FLOW RATE CONTROL: the user sets the flow rate (the command is a flow rate) and the system continuously adapts the applied pressure difference so that the flow rate Q meets its target.

You can refer to this video to learn more about pressure-driven flow control

<https://youtu.be/niWfINDUub4>





1 MUX Distribution

12/1 bidirectional microfluidic valve

The rotary valve is the second highlight of the setup. This instrument can switch between up to 12 liquids. It enables the injection of 1 sample into 12 outputs or 12 samples into 1 output. Used with the pressure controller, it allows the sequential injection of solutions in a microfluidic system.

1 Manifold

It enables merging or splitting microfluidic liquid or gas lines. When a port is not used, it has to be plugged with a plug fitting (included with the manifold) to avoid any leakage. Several manifolds can be added in series to merge or split as many lines as required.



1 Flow rate sensor (optional)

The flow rate sensor measures the flow rate of the liquid going through the sensor.

Used with the pressure controller, it enables the monitoring and control of the flow rate. Different flow sensors are available in the Elveflow product range corresponding to different ranges of flow rate detection. Your choice of flow sensor will depend on the requirements of your experiment including injected volumes, flow rates, types of liquids...

For example, if multiple solutions with various properties are involved, working with a coriolis force based flow sensor (BFS) is highly advised to avoid laborious calibration steps thanks to its compatibility with nearly any solution.

Feel free to reach out to us at contact@elveflow.com if you have any questions.



Reservoirs

All the liquids are stored in reservoirs. A different reservoir is required for each solution.

Six different volumes of reservoirs from Eppendorf (µL) to bottles with GL45 caps (L) are available, allowing versatility in your volume operations.



Starter Pack Kit

All the following items are included in our Starter Pack. It will help you to easily set up your system and connect all its components together:

Refer to **appendix 1** for a visual guide to connections

1 PTFE Tubing 1/16" OD 1/32" ID

The liquid passes through the tubing to go from one component (such as reservoirs, MFS, microfluidic device...) to another. The internal diameter equals 1/32" or 794 µm. The outer diameter equals 1/16" or 1590 µm. Other tubing sizes are available on request.





Fluidic resistance tubes

The fluidic resistance are tubes with small diameters, which will help to achieve better and more stable control of the flow rate. The higher the resistance, the higher the pressure (from the OB1) to get a given flow rate. Unions (see connection kit below) are necessary to connect the microfluidic resistances with the rest of the system.



To learn more about resistance

<https://support.elveflow.com/support/solutions/articles/48001142611-set-a-flow-control-feedback-loop-resistance-and-pid-tuning>

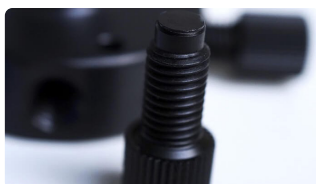
1 Microfluidic connection kit

These pieces are mandatory to connect the different components with the tubing.

Refer to **appendix 1** for a visual guide to connections



Union



Fittings 1/4"-28 flat-bottom fittings to block one or more ports of the manifold



Fittings for the pneumatic tubing



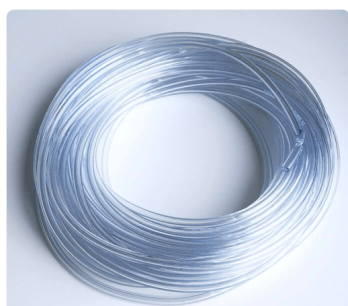
Fittings 1/4"-28 to 1/16" OD fittings for the tubing



Luer lock filter



Pressure source connection kit



Pneumatic tubing

This tubing is used to connect the OB1 pressure outputs to the reservoirs.

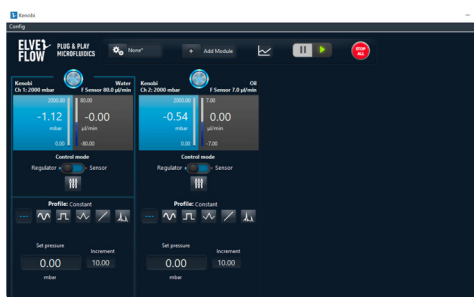


1 air filter

The air filter is placed between the pressure generator (e.g. compressor) and the OB1 inlet. It keeps the OB1 clean from dust and humidity.

1 Tubing cutter

The cutter is useful to properly cut the tubing (burr-free perpendicular cut). It helps to prevent leaks in your setup.



ESI software

All the elements of the system can be controlled thanks to the ESI software: the OB1 parameters, the rotation of the valve, etc.

Its interface allows you to easily set the pressure or flow rate you wish to apply to the system, as well as select the rotary valve port to inject the selected liquid.

It also includes a built-in sequencer for easy system automation. The provided set of SDKs in C, LabVIEW, Matlab and Python can be used for further automation and integration into larger systems.

**DOWNLOAD THE LATEST VERSION
OF ESI SOFTWARE FOR FREE**

Refer to **appendix 4** for more information on automation



The pressure source is an option to the pack.

The OB1 should be connected to a pressure supply. The optimal pressure source has to deliver a pressure higher than the maximum pressure of your OB1 channel (or at least 1.5 bar), and a maximum of 10 bar.

For example, if you are using a 0-2 bar pressure channel, working with a pressurized source delivering 2,5 bar is required to cover the whole pressure range. Refer to the [OB1 user guide](#) for more information and feel free to contact us at contact@elveflow.com to help you pick the optimal pressure source for your system and provide the pressurized air source if needed.



Before starting the experiment

Before starting the experiment

1

To begin with, make sure you have installed the software.



You can refer to this video

<https://www.elveflow.com/microfluidic-applications/setup-microfluidic-flow-control/ob1-pressure-controller-install-box/#esisoftwareinstall>

Use **version 3.05.00** or above to work with the **MUX Distribution 12**

REFER TO ANOTHER USER GUIDE



ESI software user guide | **Installation**

User safety



Before manipulating any instrument, **please read its specific user guide**.

They can be downloaded from the [support section](#) on our website.

Each user guide describes **good practices for handling Elveflow instruments**.

2

Add the OB1 on the software and calibrate it

To be calibrated, the OB1 must be connected to a pressure source, to the computer with the ESI software, and plugged to the power supply.

Once the OB1 is correctly connected, switch it on by pressing on the front panel button, **close all its channels** using the provided plugs, add it to the software and perform its first calibration.



Don't forget to **set up the provided filter** between **the pressure source** and the **OB1**.

REFER TO ANOTHER USER GUIDE



Refer to the OB1 user guide for detailed instructions | **Instrument connection**



You can refer to this video

<https://www.elveflow.com/microfluidic-applications/setup-microfluidic-flow-control/ob1-pressure-controller-install-box/#firstcalib>

It is important to calibrate the OB1 the first time it is added to the ESI software. Once done, it is not necessary to calibrate it before each experiment, unless you observe pressure drifts or if you change the pressure source.

Once your OB1 is calibrated, you can connect it to any other component of the system to perform experiments.

See **Appendix 1** for details on connecting each part of the microfluidic system

3

Add the MUX Distribution to the software

The MUX Distribution must be connected to the computer using the provided **USB cable** and **plugged to the power supply** before use. Switch on the MUX distribution.

You can then **add it to the software** by clicking on the Add instrument button > choose **MUX Distrib/Inj/Rec** > select **your instrument**.

You should then give it a name, select the MUX version (MUX Distribution 12) and **press OK** to save changes. Your MUX should be in the list of recognized devices.

REFER TO ANOTHER USER GUIDE

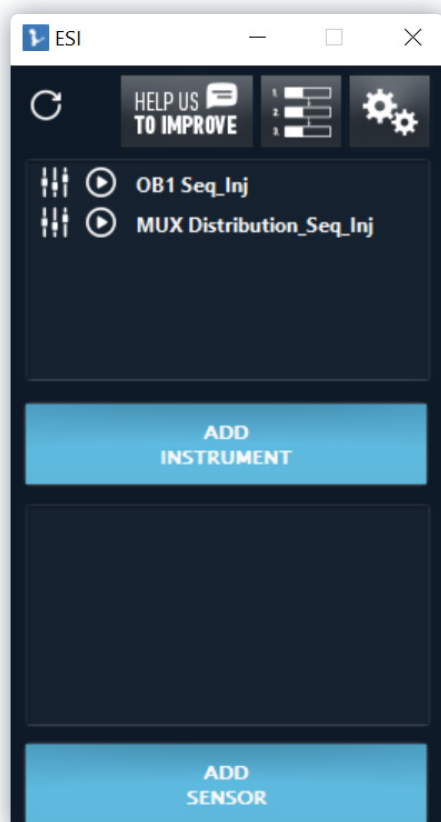
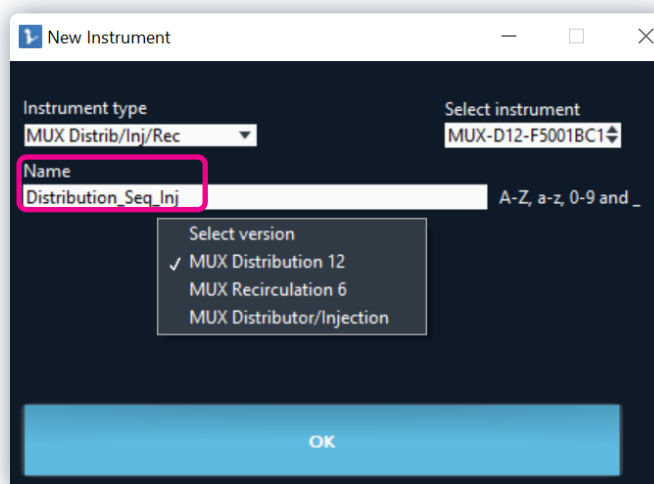
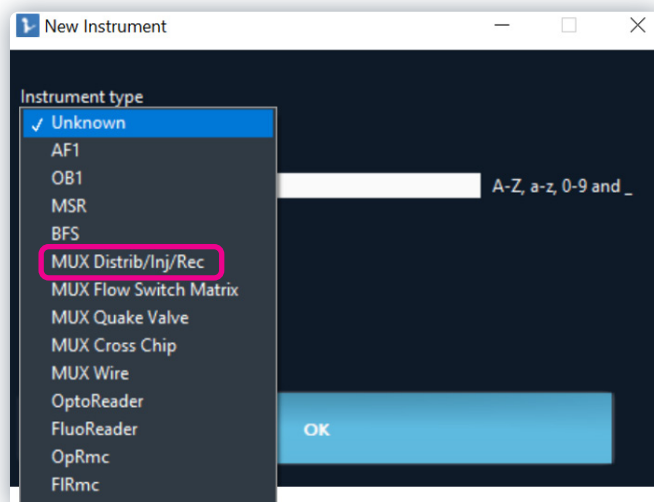
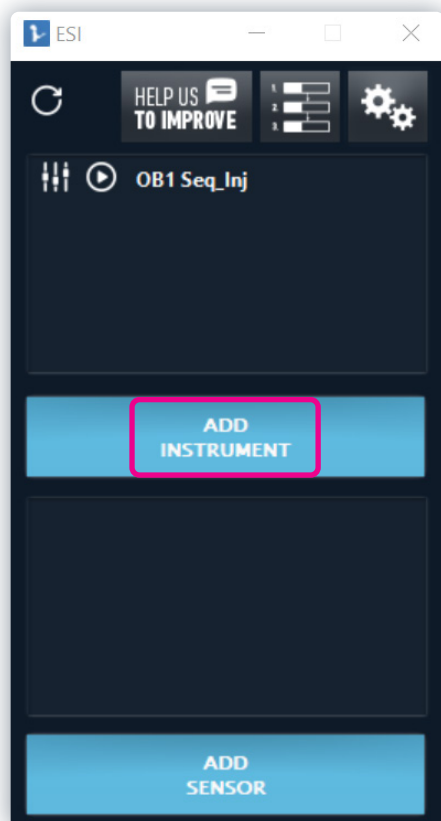


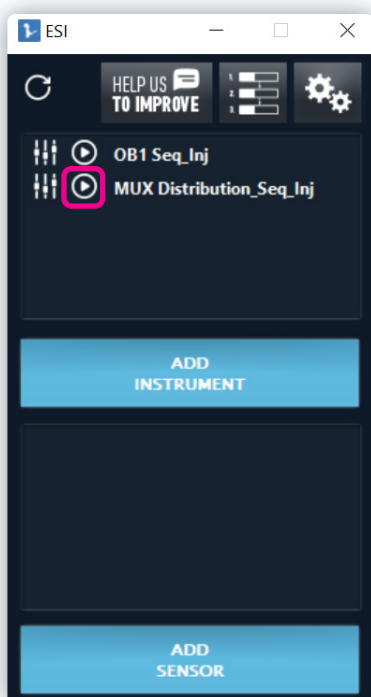
MUX Distrib user guide | **Installation & use**



ESI software user guide | **Add a new instrument**

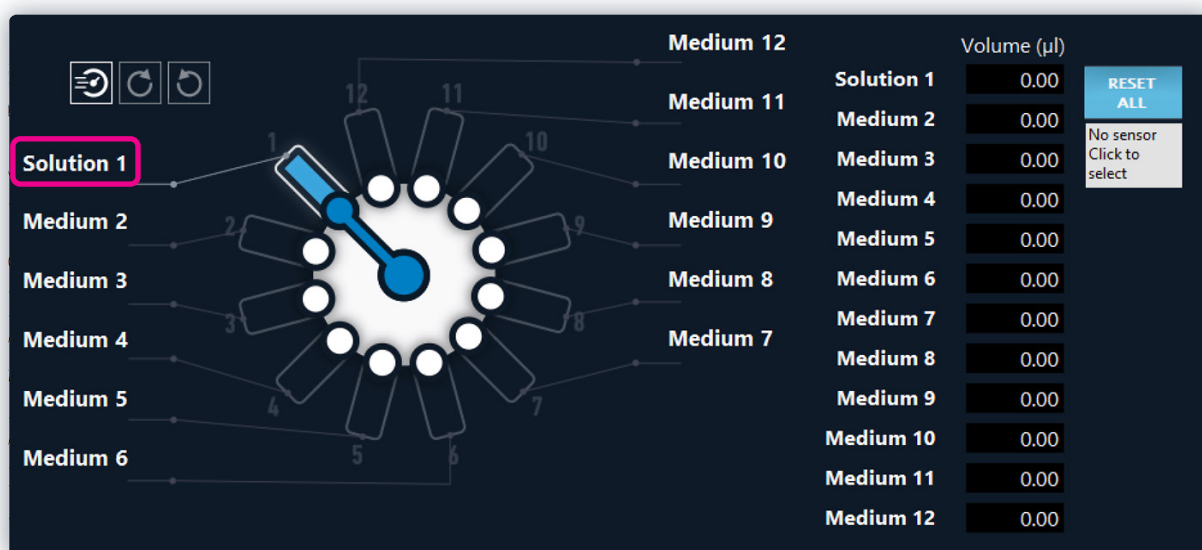






When you open the **MUX Distribution window** (arrow next to the instrument in the main window of the ESI), a noise might be heard.

To simplify your workflow, it is advised to rename the ports in the MUX Distribution window according to the solutions they are connected to. This is especially recommended if you are working with **numerous samples**.



Let's start injecting different solutions sequentially!

Let's start injecting different solutions sequentially!

1 Setup installation

Refer to **appendix 1** to see how to connect all the components together

Preparation of the reservoirs

Prepare the reservoirs filled with different solutions and connect the supplied 1/16" OD tubing and the coil tubing 4mm OD to the reservoir cap.

For more details, refer to the video [Connector for the OB1](#).

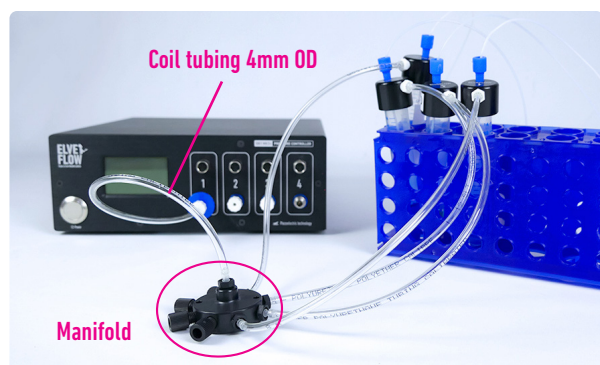
REFER TO AN ONLINE RESSOURCE



The Elveflow reservoirs connection instructions are covered by the "Quick Start Guide" | Elveflow microfluidic reservoirs assembly instructions
<https://support.elveflow.com/support/solutions/articles/48000983958-ob1-setup-installation-instructions>



Connection to the manifold



To start with, connect **the central port of the manifold to the OB1 channel output**, using the 4mm OD coil tubing. Then, connect each of the reservoirs to one of the outer ports of the manifold using 1/4"-28 thread to 3/32" OD barb fittings (black and transparent fittings).

TIP

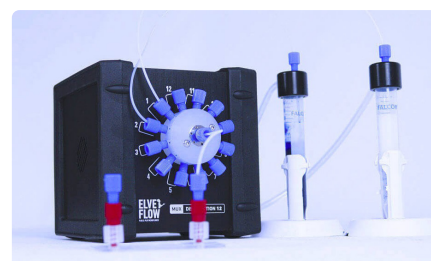
If you are not using some ports of the manifold, close them using microfluidic plugs PFA 1/4"-28 flat-bottom fittings provided with the manifold.

MUX Distribution connection and preparation

Connections

1

Connect all the reservoirs containing the solutions to the outer ports of the MUX Distribution by using the supplied 1/16" OD tubing and the 1/4"-28 to 1/16" OD fittings. At the outlet of the MUX Distribution, connect one piece of the supplied 1/16" OD tubing using the 1/4"-28 to 1/16" OD fitting and place the other end to a waste reservoir.



TIP

Connect the solutions in the order of injection to optimize the rotation of the valve. Three options for the motion are available: clockwise, counterclockwise and shortest.

TIP

In order to simplify screwing the fittings to the MUX Distribution, don't hesitate to move the instrument in order to face the hole with the fitting.

TIP

It is advised to plug the unused ports of the rotary valve with 1/4"-28 flat-bottom fittings provided with it.

Remove air from the tubing

2

On the MUX Distribution window, select port #1 and set a low pressure to fill the tubing with the liquid (e.g 50 mbar). Once liquid gets out to the waste, air is completely removed from the system and you can switch to the next liquid.



If no liquid comes out of the MUX Distribution, refer to the **Troubleshooting** section

TIP

To remove bubbles from the tubing and fill the tubing faster, it is recommended to fill the tubing from the reservoir to the MUX Distribution outlet by setting a low mbar command on the OB1. When the interface reaches the tubing extremity, set the pressure to zero. It is quicker to fill if you have the exit of the MUX directly over a waste rather than connected to all the modules that follow

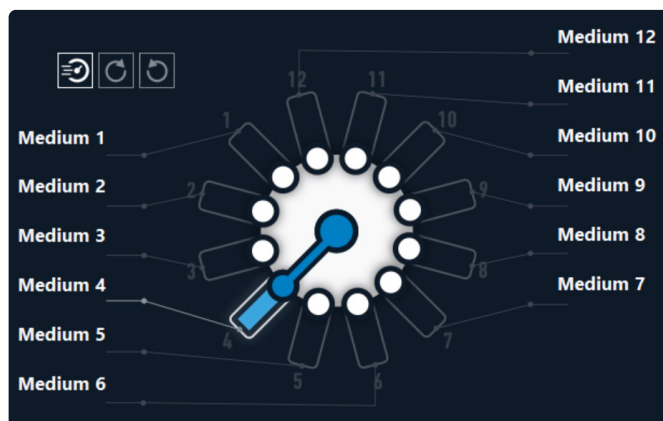
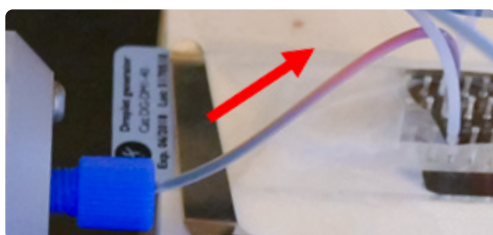
3

Repeat step 2 for all the different solutions connected to the MUX Distribution by selecting the corresponding port on the MUX Distribution window. A noise is heard during the rotation of the valve.

REFER TO ANOTHER USER GUIDE



MUX Distrib user guide



TIP

It is also possible to introduce a 3/2 valve at the outlet of the MUX Distribution in order to flush the MUX from air or a waste solution. Refer to the "Going further" section, appendix 7, for more information.

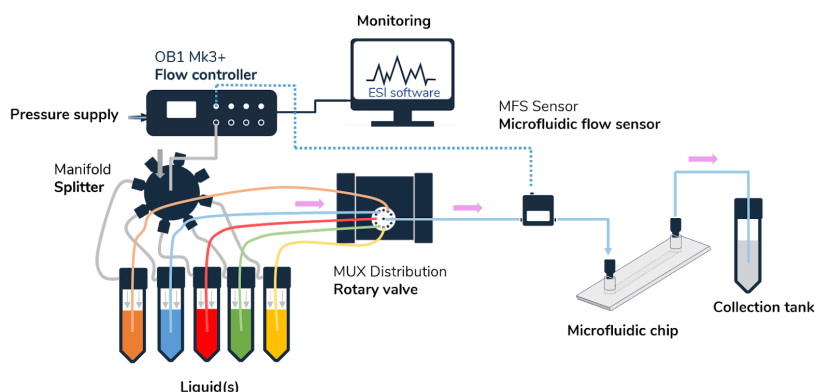
Flow sensor connection (optional)

For **flow measurement or control**, connect the flow sensor between the MUX Distribution and the microfluidic device



Refer to the **MFS user guide** for more details

A resistance tubing can be added to the system right after the flow sensor. The use of the flow sensor to control the flow rate is described in the "Going further" section, **appendix 2**.



Device connection

Connect the tubing of the MUX Distribution outlet (or the flow sensor outlet if you plan to control or monitor the flow rate) to the corresponding inlet of your device.

Connect another tubing to the outlet of the device and place the other end of the tubing in a waste reservoir.

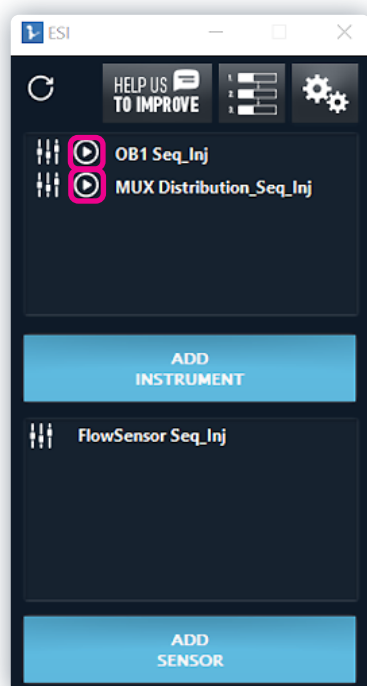
TIP

To keep reducing the formation of bubbles and keep shortening the initial filling time, it can be now useful to fill the tubing from the MUX Distribution outlet to the device with liquid before connecting the device. To do so, repeat the same steps as stated earlier to remove the air from the tubing arriving at the MUX Distribution. Set a low mbar command on the OB1 and watch the liquid interface going through the tubing, set the pressure to zero when the interface reaches the tubing extremity then connect the extremity to the device.

2 System control: sequential injection

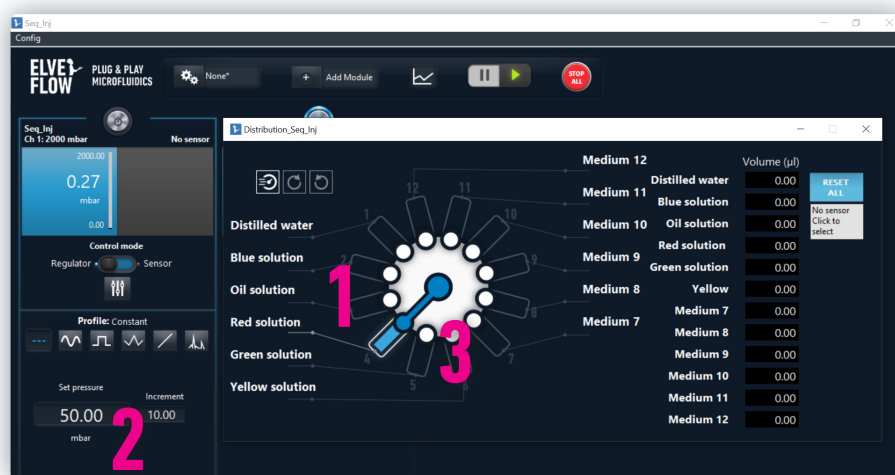
Now that **all the reservoirs are filled with their respective liquids** and all the components are connected together (OB1, manifold, reservoirs, MUX Distribution, device), **switch on** the OB1 pressure controller and the MUX Distribution and launch the ESI software.

Open the OB1 and the MUX Distribution windows in the ESI as introduced in the “**Setup Installation**” section.



TIP

If you have previously connected the instruments to the ESI software, you might just need to refresh the main window of the ESI in order to make the instrument appear.



1. Select a port on the MUX Distribution window. In the example, the red solution is injected into the system.
2. Set the channel pressure to a **given value** until the microfluidic device is filled. The pressure is set to 50 mbar in the example.
3. To change the solution injected to the device, go back to the MUX Distribution window and **select another port**. The valve rotates (a noise can be heard), another solution is selected and injected.

CONGRATULATIONS, YOU HAVE SEQUENTIALLY INJECTED DIFFERENT SOLUTIONS!

TIP

For a faster switch between liquids, place your device as close as possible to the MUX Distribution. This also allows more accurate control of the injected volumes with lower sample volumes

Browse the appendices to discover many useful tips and guides for taking full advantage of the versatility offered by this system, e.g. control by flow rate (Appendix 2), or achieve precise control over an injected volume (Appendix 3).

TIP

If the solutions are not injected in the system in the order they are connected to the rotary valve, be aware of possible cross contamination. In fact, when switching between two non consecutive ports and depending on the pressure/flow rate applied, the valve takes a small volume of all the solutions connected to the ports in between. As a result, the first volume going out of the valve is a heterogeneous solution. Refer to the “Going further” section, appendix 7, to find information on how to solve this issue by adding valves to the microfluidic line.

3 Technical specifications

The sequential injection system is designed to easily inject different solutions in a microfluidic device. User friendly and versatile, the system's main characteristics and options are summarized in the following table.

SEQUENTIAL INJECTION SYSTEM		SPECIFICATIONS	
Range of pressure	0-7 bar		
Range of flow rates	from 70 nL/min to 5 mL/min	from 1.6 µL/min to 500 mL/min	
Type of liquids	With MFS: 2 calibrations (water and IPA)	With BFS: any liquids	
Switching time	With pressure control: 150 ms	With flow control: 1-2 s (depending on your system's parameters)	
Number of solutions	from 1 to about 150		
Wetted materials	PCTFE and PTFE		
Operating temperature	5 °C to 40 °C		
Operating humidity	20 to 70 % condensing		

TIP

Some important but quick variations of the flow rate are observed each time the MUX Distribution valve rotates, especially when the flow rate is steady. To learn more about it, refer to the “Troubleshooting” section.

TIP

In order to decrease the impact of the mechanical switch of the rotary valve on the stability of the flow rate, we advise to set the pressure (or the flow rate) back to zero, then apply the switch of ports on the MUX Distribution and set back the pressure (or flow rate) to the targeted value. Refer to appendix 9 of the “Going further” section for more tips.

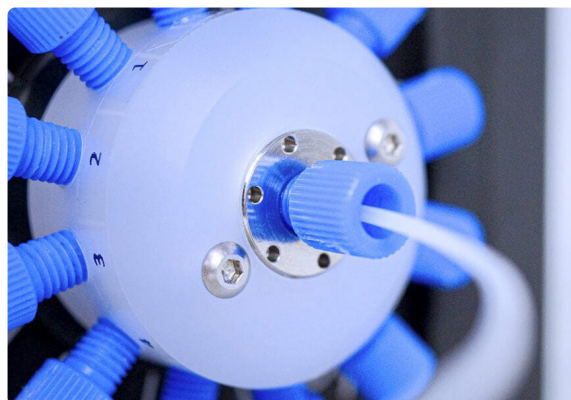
4 After the injection

Once your experiment is finished, make sure to empty the tubings and clean the rotary valve by flushing at least distilled water in all the lines of the valve, followed by air drying. This procedure is sufficient to clean the tubings and valve from **aqueous or oil solutions**.

REFER TO ANOTHER USER GUIDE



MUX Distrib user guide | for more information on cleaning procedures **if you work with other solutions**



TIP

To dry the rotary valve, connect a reservoir filled with air to the valve. Pushing air allows you to remove the remaining solution stuck in the valve. You can also work with gas and have reservoirs filled with different types of gas.



Troubleshooting & Going further section

Troubleshooting

The MUX Distribution inlets/outlets leak

The reason is probably that the fittings are not tightened enough on the MUX Distribution valve.

Before applying a pressure to the microfluidic line, make sure to have tightened the fittings well enough. Be careful not to over tighten the connector though to avoid any long term damage.

No solution comes out of the MUX Distribution

One reason could be that the port selected on the MUX Distribution is not the right one. First, check the selected port on the software.

If the problem remains, the issue is likely coming from the pressure (or flow rate) target set on the OB1 pressure controller window. The target is probably not high enough for the fluidic [resistance](#) of the system (or the fluidic resistance is too high, it depends on the point of view). Disconnect the tubing coming out of the MUX Distribution from the rest of the microfluidic line and check if the fluid flows correctly out of this tubing.

If the problem remains, it could be because the communication between the instrument and the software is interrupted. To restore the communication, you can :

(1) select another port and come back to the previous one,
(2) close the MUX Distribution window, open it again and select the port needed
or (3) click on the “refresh” button on the main window of the ESI.
A noise should be heard during rotation if the MUX valve is rotating correctly.

There are big jumps in the flow rate when the valve is rotating

When switching the valve, the flow rate first decreases when the flow is blocked. To compensate for this stop, the pressure increases and makes the flow rate overshoot the set value before going back to this value in less than 1s. So the observation of big jumps in the flow rate when the valve rotates is normal.

To learn how to lower the impact of the mechanical switch on the flow rate, refer to **appendix 9** of the “Going further” section.

There are bubbles in my tubing

One reason could be that some air was left in the tubing when you connected a new reservoir to the MUX Distribution.

If it is the case, disconnect the tubing going out from the MUX Distribution from the device or the flow sensor (depending on your setup), apply a low pressure and wait until the air is out. Once the solution is dripping, you can connect the tubing to the microfluidic line again. You can also refer to the MUX Distribution and preparation section for more information or add a bubble trap in your system if the problem is recurrent.

The flow rate measured by my MFS suddenly drops for a short duration

The reason is probably that you have air bubbles in your system. When an air bubble passes through the MFS, the sensor measures a zero flow rate as long as the bubble is inside. To get rid of air bubbles, just wait for it to get out by maintaining a constant pressure. The initial filling of the system is critical to avoid bubbles.

The flow rate is not stable

If the flow rate is not stable on the set value, first check that you have not forgotten to add some resistance tubing in the fluidic system. If it is not the cause of the problem, you should consider changing the values of the P and I parameters. The lower the value of P, the more stable the flow rate.

Appendix 2 of the “Going further” section details this solution.

You can also increase the microfluidic resistance. Refer to the “Going further” section, appendix 2 as well, for more information.

The OB1 is very noisy

If your OB1 starts making loud noise, it is probably because of a leak in your system: the pressure source tries to permanently compensate for the lack of pressure due to the leak. Check all the connections of the system (using teflon tape is often useful to avoid leaks on fluidic and pneumatic connections).



This application note is dedicated to the solution

<https://support.elveflow.com/support/solutions/articles/48001143505-fix-ob1-continuous-noise-or-a-leak>

There is dust in my tubings or device/my microfluidic line is clogged

In case your microfluidic line starts to clog (dust, particles, etc), try to increase the pressure (or flow rate) to expel the dust from the line. If it does not work, you should change the resistance tubing. As its diameter is smaller than the rest of the tubings, it is generally the first to be clogged. Another solution might be to change the device you are working with. The presence of dust in the line should not be ignored. The flow rates can be impacted and the volumes injected will not match the expectations. Working with “clean” solutions and reservoirs is essential to prevent clogging. We highly advise to filter solutions before using them in a microfluidic line.

Going further

The **sequential injection pack** is very flexible, and it might be beneficial to further customize your setup to perfectly fit your requirements.

For example, you might want to work with a specific system, or add more solutions to the setup to increase the versatility of the injection, or perform a complete automation of your injection system.

Be aware that even though **it is not hard to adapt your setup to new experimental conditions**, there are always a few elements to fine-tune to make sure your experiment works well!

The following appendices are dedicated to the improvements you could make to your system to further boost your experiment!

Appendix 1

Microfluidic connector guide

Connect the OB1 to the pressure source



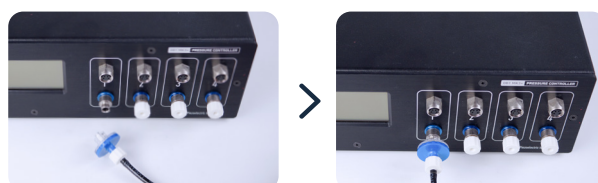
Connect your OB1 pressure controller to external pressure supply using pneumatic tubing

REFER TO AN OTHER USER GUIDE



Refer to the OB1 user guide for detailed instructions

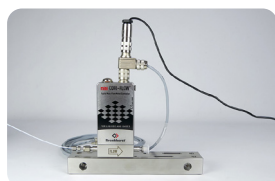
Connect the reservoir to the OB1 pressure outlet



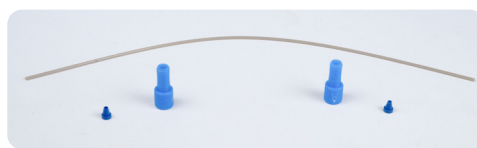
Prepare the MFS



Prepare the BFS



Prepare the resistance



Connection MFS - Resistance



Connection Resistance - Tubing



Connection Tubing - MFS



Connection tubing - valve



Connection tubing - manifold



Flow rate control

Appendix 2 Flow rate control

This section explains how to **directly control the system in flow rate**, through a feedback loop between the pressure controller and the flow sensor. In **feedback control mode** ("Sensor" mode), you can set a target flow rate, and the inlet pressure will automatically adapt to reach it. The feedback loop relies on a PID algorithm. The D parameter is preset but the gain parameter (P) and the integration parameter (I) can be tuned.

We advise you to use the "PI Basic" algorithm that will work for most applications. The user can use the autotune module to automatically find P and I parameters, or set these parameters manually to better match the requirements in terms of responsiveness and stability.



WHY CONTROL THE FLOW RATE RATHER THAN THE PRESSURE?

Requesting a flow rate instead of imposing a pressure has three main advantages and is necessary if you plan to control the injected volumes of your solutions:

- **The flow rate stays constant throughout time**, even though the experimental conditions slightly change. For example, if a bubble or clogging occurs, if the temperature widely changes..., the system compensates and maintains a constant flow rate.
- **It compensates for hydrostatic pressure changes**. The hydrostatic pressure is the pressure exerted by a fluid column on a surface. When the tube inlet is at the bottom of the reservoir, the hydrostatic pressure depends on the height of liquid between the liquid/air interface and the bottom of the reservoir. During the experiment, when the level of fluid in the reservoir decreases, the hydrostatic pressure follows the same trend (around 1 mbar per cm of water). As a result, the pressure at the inlet of the fluidic system will also decrease, and so will the flow rate. In flow rate control ("Sensor" mode), the pressure is adjusted to compensate for changes in hydrostatic pressure and hence maintain a constant flow rate.
- Even if your setup changes from one experiment to another, **working with flow rates will allow you to more easily reproduce your experiments**.

STEP 1

To **control the flow rate**, first add a flow sensor to your main microfluidic line. We recommend that you place it between the MUX Distribution and the device. If it is not already done, refer to the section **Setup installation**.

Add the flow sensor on the software by clicking on the **"ADD SENSOR"** button. Choose the type of sensor (analog or digital) and the instrument and channel it is connected to. It is advised to give the MFS appropriate name.

REFER TO ANOTHER USER GUIDE

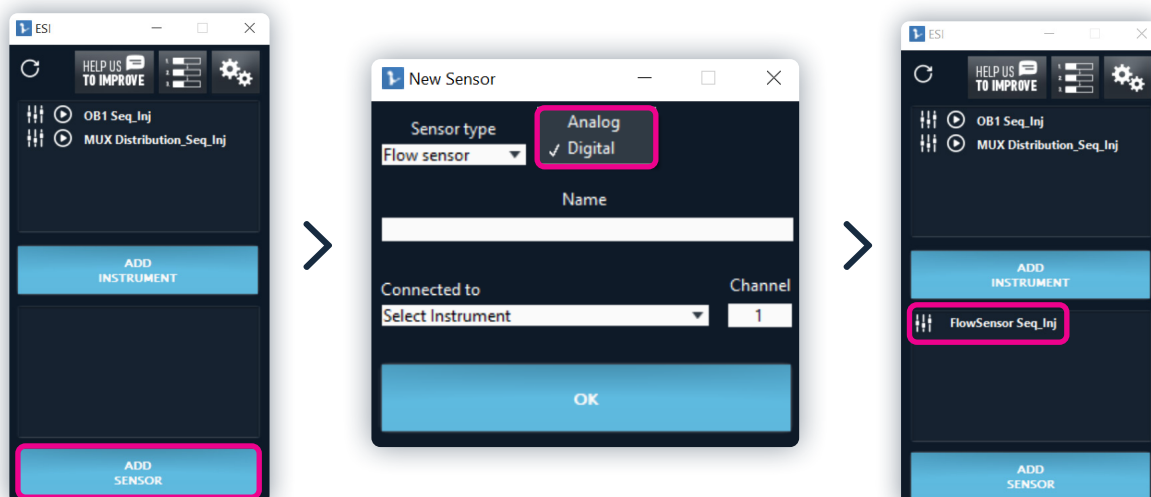


MFS user guide | **Electronic connection**



ESI software user guide | **Add a sensor**

PDF



TIP

If you plan to inject liquids with properties different from aqueous solution or IPA, a calibration is required following the calibration procedure. Another option is to use a BFS sensor as this sensor works with any liquids and does not require any calibration. To learn more, you can refer to "[Calibrate MFS flow sensor with any liquid](#)".



BFS: principle, advantages and specifications

The Bronkhorst Flow Sensor (BFS) is based on the Coriolis technology and presents unique benefits for your microfluidic experiments.

- **Versatility:** it works with any liquid and doesn't require any calibration to ensure its compatibility;
- **High accuracy:** up to 0,2% of the flow rate;
- **Broad flow rate range:** a single sensor measures from $\mu\text{L}/\text{min}$ to mL/min (several ranges available from 1.6 $\mu\text{L}/\text{min}$ to 500 mL/min);
- **High flow rates accessible:** up to 500 mL/min .



Be aware that this flow sensor is added to the software **as an instrument** and not as a sensor. To learn more, refer to the [dedicated webinar page](#) and the [Bronkhorst website](#).

WHY IS IT NECESSARY TO CALIBRATE THE MFS FOR LIQUIDS OTHER THAN WATER AND IPA?

MFS flow sensors measure the flow rate of a liquid by locally warming it (max 0.5 °C) and by measuring the temperature drop across the sensor capillary. For comparison, the BFS sensor works with the Coriolis force. The relationship between temperature and flow rate is highly dependent on the physical properties of the liquid passing through the MFS, which is why calibration is required. Two calibrations are natively implemented in the digital MFS : water and Isopropyl alcohol (IPA). The water calibration is suitable for all aqueous solutions. The IPA calibration is appropriate for use with liquids of any hydrocarbon chain length, with an additional linear adjustment through calibration.

A [specification table](#) gives all the corresponding flow rate ranges depending on the calibration for all the MFS flow sensors.



STEP 2

Once the MFS flow sensor is hooked up directly to the OB1, you can visualize the flow rate in your microfluidic setup.

Go to the OB1 window.

On the right part of the channel (grey rectangle), the flow rate is now indicated. Right above, the name of the flow sensor connected to the pressure channel appears.



To learn more about the different working principles of flow sensors

<https://www.elveflow.com/microfluidic-reviews/microfluidic-flow-control/microfluidic-low-flow-liquid-flow-meters-a-review/>

STEP 3

The next task is to check the fluidic resistance of the setup and adapt it if necessary.

The addition of resistance tubing might be necessary in order to adapt the overall fluid resistance of your setup. If this is the case, the length and diameter of the resistance tubing need to be determined.

TIP

If you change the microfluidic device, flow sensor or the liquid in your setup, the fluidic resistance might be different. If so, you will have to adapt the length and/or diameter of the resistance used in the system, so that the range of the pressure controller matches the range of the flow rate sensor you work with.

TIP

When the fluidic resistance of the system is well dimensioned, it is possible to span the whole range of flow rates of the flow sensor with the range of pressure of the OB1, avoiding saturation of the flow sensor.

TIP

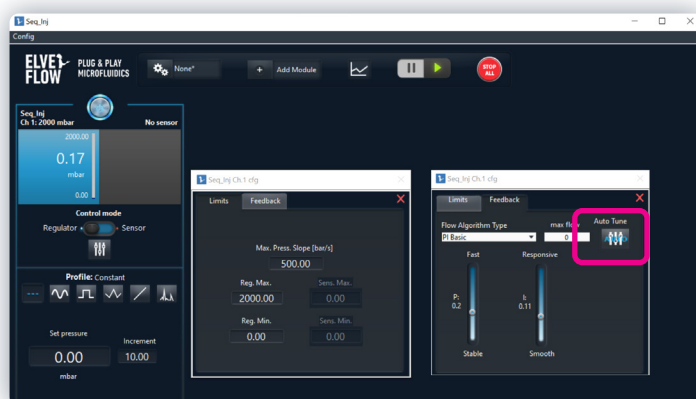
The resistance should always be placed downstream of the flow sensor (between the sensor and the device) to get the most stable measurements.

STEP 4

The last task is to **tune the P and I parameters** in order to **adapt the stability and responsiveness** of the flow rate to your requirements. Several algorithms are available.

Elveflow recommends using the "PI Basic" algorithm which gives good results for most of the applications.

First, we advise you to use the autotune of the ESI software. This option suggests a couple of P and I parameters giving a good balance between stability and responsiveness.



TIP

If you can't get the responsiveness and stability you aim at with the "PI Basic" algorithm, you might use the "PI large reservoir" algorithm instead. This second algorithm is more adapted for large volumes (lots of air).

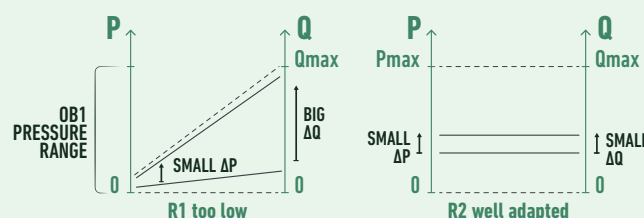


WHY IS MICROFLUIDIC RESISTANCE IMPORTANT?

Adding the right amount of fluidic resistance in the fluidic path is important as it is the way to finetune the system's whole resistance R to obtain the best performance in terms of flow rate control. It allows to adjust R so that the range of accessible ΔP (given by the range of the OB1 channel output) matches the range of the flow rate sensor used. Doing so, the system uses the whole dynamics of both the sensor and the regulator to control the flow rate.

For instance in a system (with a 2 bar OB1 channel) where $R=R1$ is too low, few mbar of ΔP generate a high flow rate that can saturate the flow rate sensor. In other terms, a small part of the accessible ΔP matches the full range of the sensor, it can lead to unstable flow control: a small ΔP variation triggers a huge flow rate variation.

The solution is to add the right amount of fluidic resistance to increase $R=R2$ so that most of the range of the pressure regulator is used. As a result, a higher pressure is required to saturate the sensor, consequently a small ΔP variation triggers a small flow rate variation and it allows to obtain a stable system.



TO LEARN MORE

<https://www.elveflow.com/microfluidic-applications/setup-microfluidic-flow-control/microfluidic-flow-restrictors/>

However, the parameters proposed by the autotune option might not be the best for your experiment as this module has a tendency to give more importance to stability over responsiveness. So, if your experiment requires greater responsiveness, you might need to fine tune the parameters by hand in order to increase this specificity.

TIP

Click and type the values of P and I and it will be easier than scrolling through values



THE NEED FOR FEEDBACK: HOW TO FINE TUNE FEEDBACK PARAMETERS (P AND I)

There are **two main reasons** that will lead to a need for readjustment of the P and I parameters:

- You would like to **monitor the responsiveness** (P), the stability of the system (I), and/or find a greater balance between both depending on your requirements.
- You **make changes to your experimental setup**, especially to the resistance or liquids, which require P and I parameters to be optimized once more.

To make the necessary adjustments, the values of the feedback parameters can be fine tuned in several ways. For example, using the autotune option with the "PI Basic" algorithm on the setup presented in this user guide, the flow rate stabilized within 30 s after values of P and I were automatically selected.

With the "PI large reservoir" algorithm, stabilization time was even longer. However, after fine tuning the P and I parameters, it is possible to balance responsiveness and stability and get to the requested flow rate of 200 $\mu\text{L}/\text{min}$ in a couple of seconds.

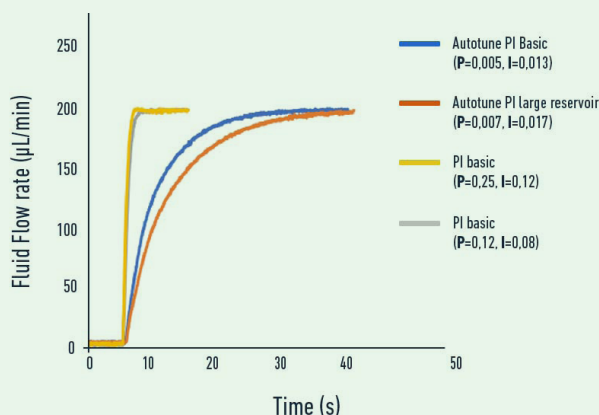
Be aware that a sudden increase of the I parameter value on the "PI Basic" algorithm while controlling the flow rate ("Sensor" mode) will induce an overflow. It is recommended to change the value of I while controlling the pressure ("Regulator" mode) and then switch back to flow rate control.

You have too much overshoot? **Decrease P!**

The system reacts too slowly? **Increase P!**

The system reacts quickly but slows down as it approaches the target? **Increase I!**

You get some instabilities or too much oscillation? **Decrease I!**



STEP 5

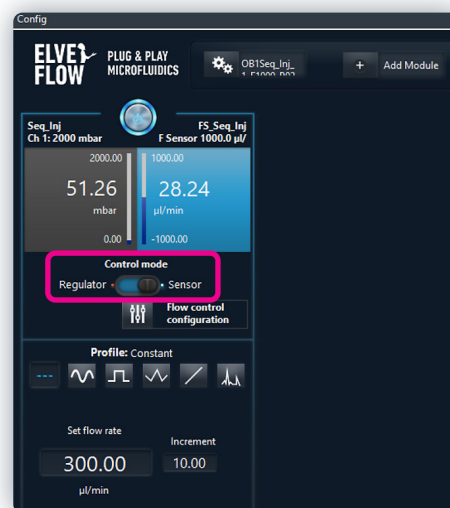
Now that the flow sensors are calibrated, the fluidic resistance and the P and I parameters of the system optimized, you are ready to **start the feedback loop of flow rate control**.



Don't forget to make sure that the whole fluidic system is filled with liquid before switching to flow rate control mode.

Choose the **"Sensor" mode** (flow rate control instead of pressure control), and set the flow rate you want to reach (you are now controlling the flow rate instead of the pressure).

Refer to the **appendix 3** if you want to watch volume injection live



TIP

While the tubing is filled with air, you should control by pressure and not by flow rate. This is because a flow sensor filled with air will not measure a flow rate, and the OB1 will keep increasing the pressure until the fluid reaches the flow sensor. Reaching the maximum pressure of the channel pressure range should be avoided. Once the whole system is filled with liquid, it is safe to switch to flow rate control.

Moreover, instabilities in flow rate will appear if air bubbles flow in the circuit, especially through the flow sensor. We advise you to add a bubble trap before the flow sensor in your setup in order to solve the problem.

TIP

The flow sensor is very sensitive to vibrations and movement perturbations so place it on a stable surface. Make sure the outlet reservoir is not totally empty and that the outlet tubing plunges into the solution. Otherwise, there will be dripping, which will affect the flow rate stability.

REFER TO AN APPLICATION NOTE



To learn more about the feedback loop, the choice of resistance tubing and the tuning of the P and I parameters

<https://support.elveflow.com/support/solutions/articles/48001142611-set-a-flow-control-feedback-loop-resistance-and-pid-tuning->

Once your experiment is finished, make sure to **empty the tubing** and to **properly clean the flow rate sensors** according to the cleaning procedures described in the Flow Rate Sensor user guides and [in the support section of the Elveflow website](#).

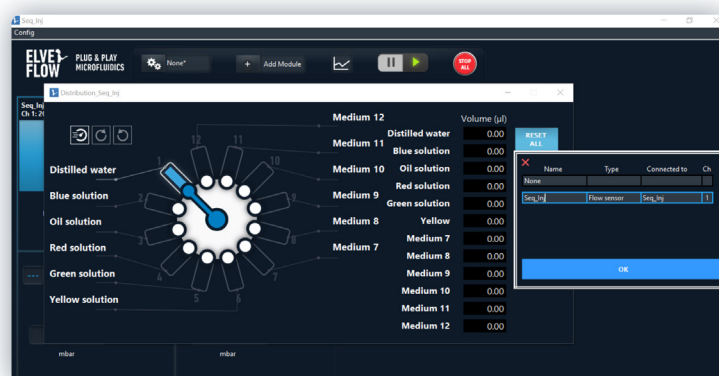
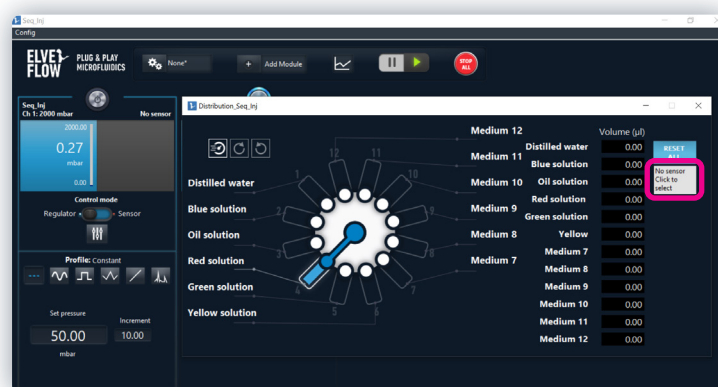
Appendix 3 Volume control - manual switching

Easily visualize the volume injected and switch between samples when a specific volume is reached

This section presents **how to follow an injected volume in real time in a microfluidic device** and **easily control sequential injections**.

This section describes **manual switching** between ports, which enables a rough estimate of injected volumes. Refer to **Appendices 4 and 10** for details on finer volume control using automated switching.

Connect the **flow sensor** to the **MUX Distribution** using the panel on the right of the screen “No sensor, Click to select”.



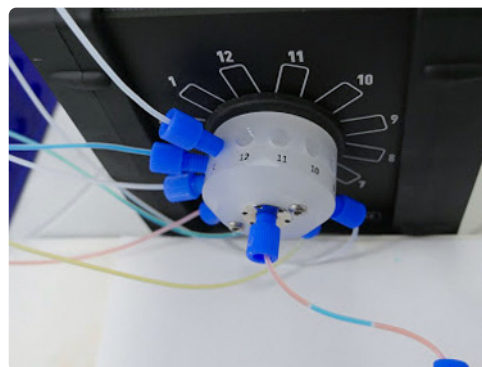
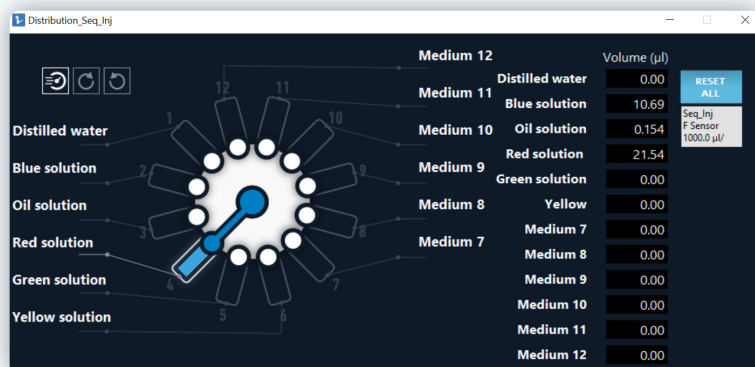
A new window appears, select the correct flow sensor and click on OK.

The flow sensor is now visible in the grey area, where “No sensor, Click to select” was previously visible.

Go back to the OB1 window and set a pressure.

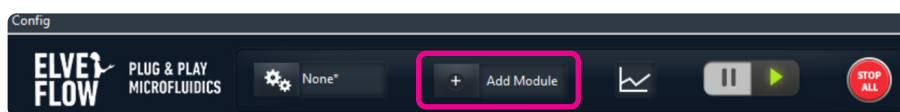
Back to the MUX Distribution window, you can follow the volume injected through the selected port. If you don't need high precision, you can manually switch between the ports when the desired sample volume has been injected.

In the example below, 10.69 μL of blue solution has been injected. The objective was to inject around 10 μL .



"Flow integration" module

This module is useful when you want to inject a precise volume of solution into a device and follow the remaining volume in the initial reservoir.

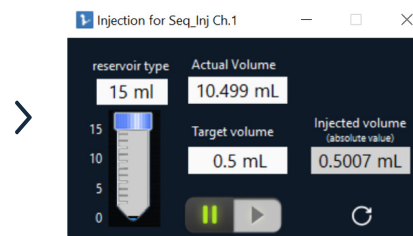
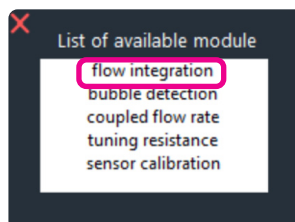


Go to the **MUX Distribution window** and select the port of the solution you want to inject.

On the OB1 control window, click on **Add Module** and select the **flow integration** module in the list that appears.

A window pops up.

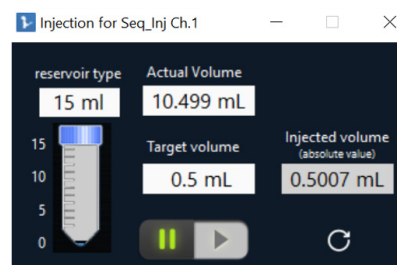
Enter the type of reservoir used, the actual volume of solution in the reservoir and the target volume and click on play.



Note that very small variations in the injected volume may be seen as a result of flow rate fluctuations, but these will be negligible with respect to the final value.

Go back to the OB1 control window and set up a pressure or flow rate.

Once the targeted volume is reached, flow will be stopped automatically.



TIP

This module can also be used to add a threshold to avoid emptying the reservoirs completely.

Appendix 4 System automation

To **easily automate experimental parameters** (e.g. pressure, flow rate, injected volumes) for fluid injection, use the integrated sequence/ automation tool of the ESI software.

To learn more about the automation of sequences using the ESI software:

REFER TO ANOTHER USER GUIDE



ESI software user guide | **P.34 sequence section**

For further integration into your system, you can also use the Software Development Kit (SDK) provided with the ESI software (available in LabVIEW, C++, Python and MATLAB).

For more information regarding the automation using SDK, refer to the SDK user guide or send us an email at **contact@elveflow.com**.

Automation using the sequencer

The sequencer included in the ESI software can be used to easily automate your sequential injection of liquids and/or control other parameters such as pressure, flow rate, volumes among others. The scheduler is built as a series of steps, each with a specific command such as load an instrument configuration or specify the length of the previous command.

Before building a sequence, it is recommended to create all the instrument configurations required for the experiment.

- 1 On the OB1 window: set the desired flow rate or pressure for your experiment on the correct channel(s), start the channel(s) (top button is coloured blue) and save the configurations by clicking on "Config".

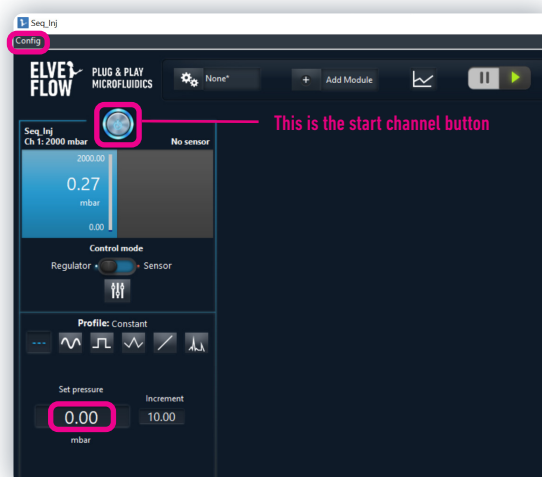
Click on the pause button to change the OB1 window settings without impacting the system: on editing mode, changes are not effective until you click on the play button again.

This way, it is possible to prepare and save all the OB1 configurations and keep a zero pressure or flow rate in the system.



For more details, refer to the **ESI software user guide**

- 2 Save a "0" pressure or flow rate configuration for all your channels. This configuration is important to stop the channel at the end of the sequence.
- 3 Step 1 can be repeated for all needed configurations.



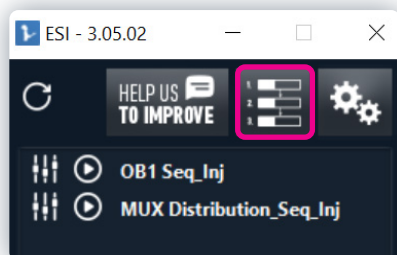
TIP

We advise you to control the flow rather than the pressure during your experiments for better accuracy. If flow is initially unstable, add a piece of flow resistance. Refer to Appendix 2 for more detailed information.

TIP

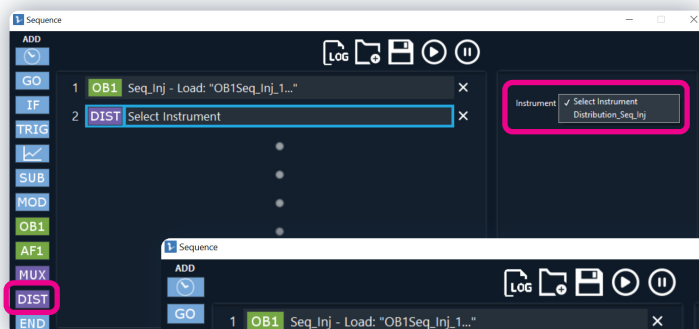
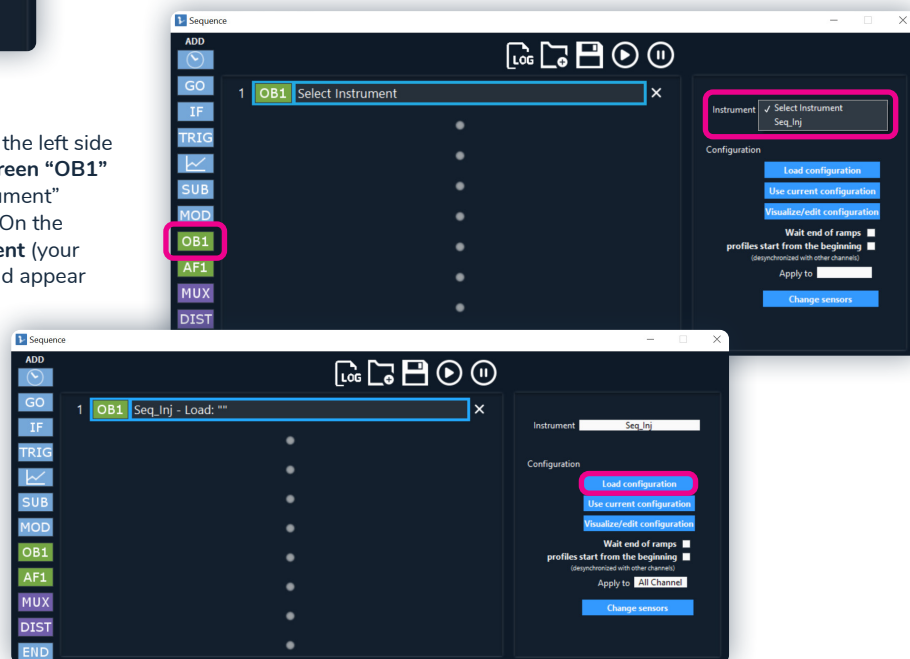
If you use different liquids and control the injection with the flow rate, be aware that the optimal P and I parameters might be different from one liquid to another. You can create different configurations for all the liquids. When saving the configurations, the algorithm parameters (P and I) will also be saved. However, when using the MFS sensors, be aware that the type of liquid the sensors are calibrated for can not be changed during a sequence. So if you work with aqueous solutions and IPA and want to automate your experiment, we recommend using a BFS sensor.

You are now ready to **start the creation of the sequence**.

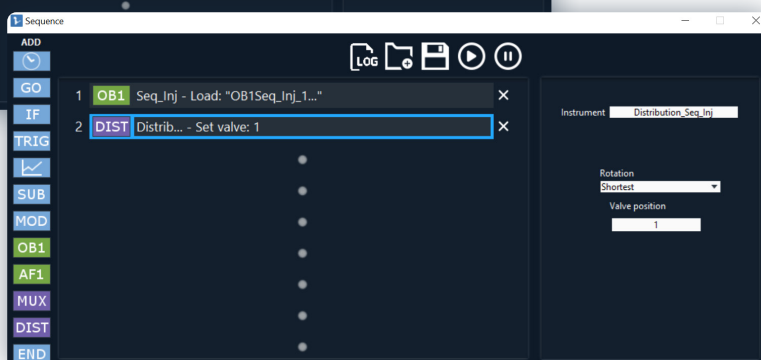


4 Click on the top middle button “**Create Sequence**” in the main window of the ESI software: a new window will appear.

5 On the sequence window: on the left side of the window, **click on the green “OB1” box** (a line “OB1: Select instrument” appears in the middle panel). On the right side, **select the instrument** (your OB1 pressure controller should appear when clicking on “Instrument”) and “load the configuration” saved previously.

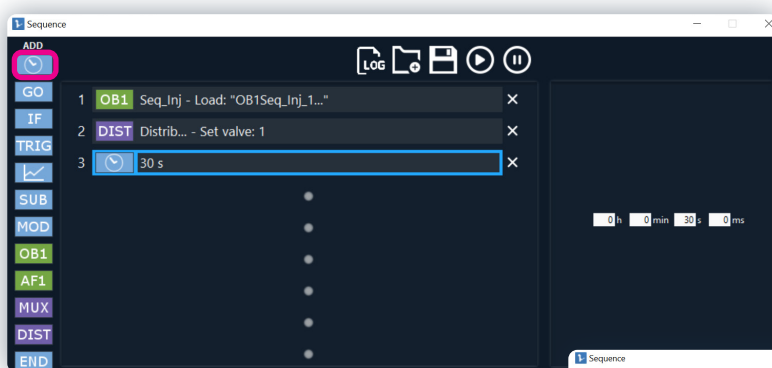


6 On the left side of the sequence window, click on the **purple “DIST” box** (a new line “DIST: Select Instrument” appears): **select your instrument** and indicate in the box “Valve position” the number of the port corresponding to the desired solution.



TIP

You can choose the rotary motion of the MUX Distribution for your convenience (shortest, clockwise or counterclockwise). We recommend choosing the shortest rotation in order to decrease the impact of the mechanical switch. To learn more about the impact of the switch, refer to appendix 9.

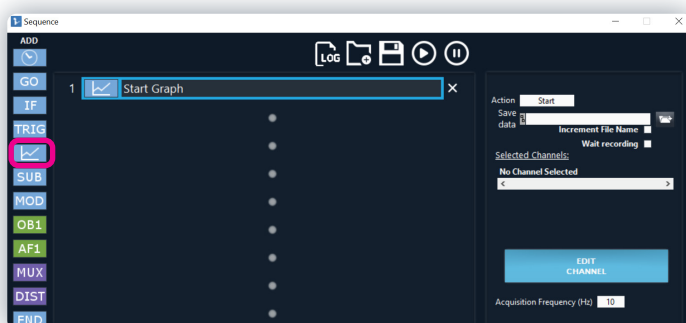
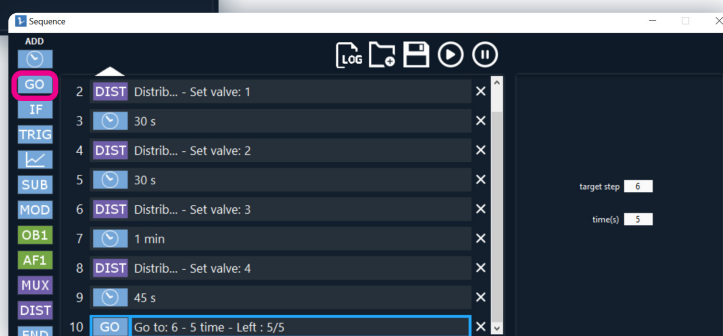


7 On the left side of the sequence window, click on the “Wait” box (clock symbol). A new line with a time frame appears: indicate the duration for which you want to inject the solution from the selected port at the chosen pressure or flow rate.

8 If you keep the same pressure or flow rate configuration, repeat steps 6 and 7 with the desired port and time to select other solutions.

9 Option: on the left side of the sequence window, click on the “GO” box.

This step will allow you to repeat your sequence in order to create a loop: choose the “target step” (6 if you want to repeat the sequence from step 6 without changing the OB1 configuration) and “time” for how many times you want this loop to be repeated.



10 Option: You can record your data (pressure and flow rate values) automatically by using the blue “Graph” box on the left of the sequence window. Click on the button, a new line appears. Choose the recording location and select the channels whose data you want to record by clicking on the “Edit channel” button. Use the same “Graph” box in order to stop the recording and only change the action on the top right from “Start” to “Stop”.

11 To end the sequence: On the left side of the window, click on the green “OB1” box (a line “OB1: Select instrument” appears in the middle panel), on the right side, select the instrument (your OB1 pressure controller should appear when clicking on “Instrument”) and “load the configuration” zero pressure or flow rate.



12 On the left side of the sequence window, click on the blue “END” box (a new line “END” appears). This means that your sequence is over.

13 Press the “Play” button at the top of the window. Your sequence will automatically run.



TIP

Steps 11 and 12 are required to stop the injection of the solutions in the device.

TIP

You can change the order of the different steps by just changing their position in the list. However, be aware that the steps you are targeting (for example when doing a loop) are not automatically updated.



TIP

You can save the sequence by clicking on the “Save” button on top of the sequence window. You can also load a previously saved sequence. To perform the loading correctly, all the instruments used when the sequence was created should be connected to the ESI software.

TIP

You can stop the sequence when it is running without waiting until the end by clicking on the “Stop” button on top of the sequence window. However, be aware that doing so, you will not stop the command on the OB1 but only stop running the sequence. The OB1 configuration will remain the one that was active when the sequence was stopped. To put the pressure/flow rate back to zero, you need to do it manually by going to the OB1 window.

Integration into larger systems using SDK or TTL

It is possible to **use SDK libraries to control the OB1 MK3+** and other Elveflow instruments with your own code and involve third party instruments such as microscopes, temperature control systems, etc. Contact us at contact@elveflow.com for more information.

REFER TO ANOTHER USER GUIDE



SDK user guide

Appendix 5 Serialization/scale up - working with more than 12 solutions

From 1 to 12 solutions

If you plan to **increase the number of injected solutions**, prepare more reservoirs and connect them to the manifold and the MUX Distribution as presented in section “MUX Distribution connection and preparation”.

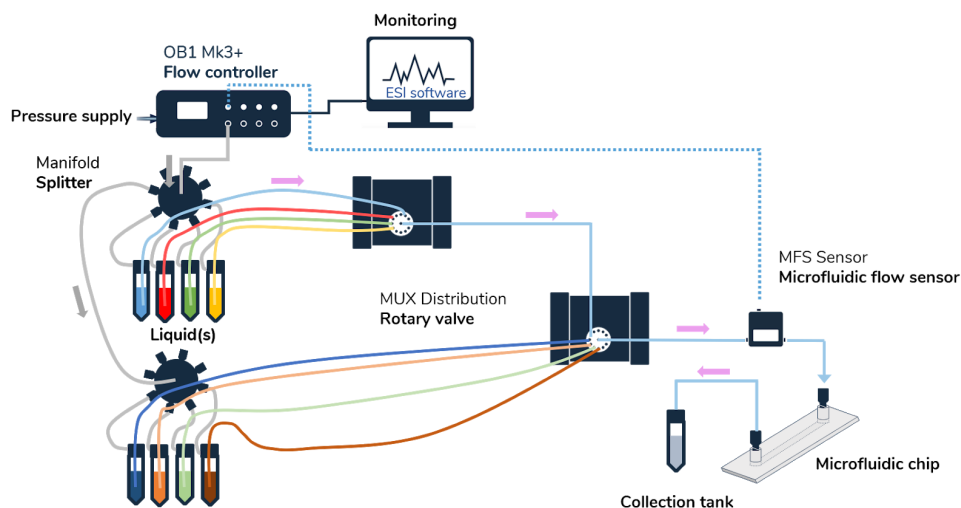
You can work with **up to 12 solutions connected to a single MUX Distribution**. When using an automated sequence, you will have to add more steps in the sequence to inject the new solutions.

From 13 to 23 solutions

If the experiment requires the injection of **more than 12 solutions**, it is possible to work with **two MUX Distributions connected in series**, meaning that the outlet of the first MUX Distribution is connected to one inlet port of the second MUX Distribution.

Using a single pressure channel from the OB1 pressure controller, it is then possible to inject up to 23 solutions.

When working with more than one MUX Distribution, you will have to open one MUX Distribution window for each instrument. We recommend giving each MUX Distribution and each solution a different name.



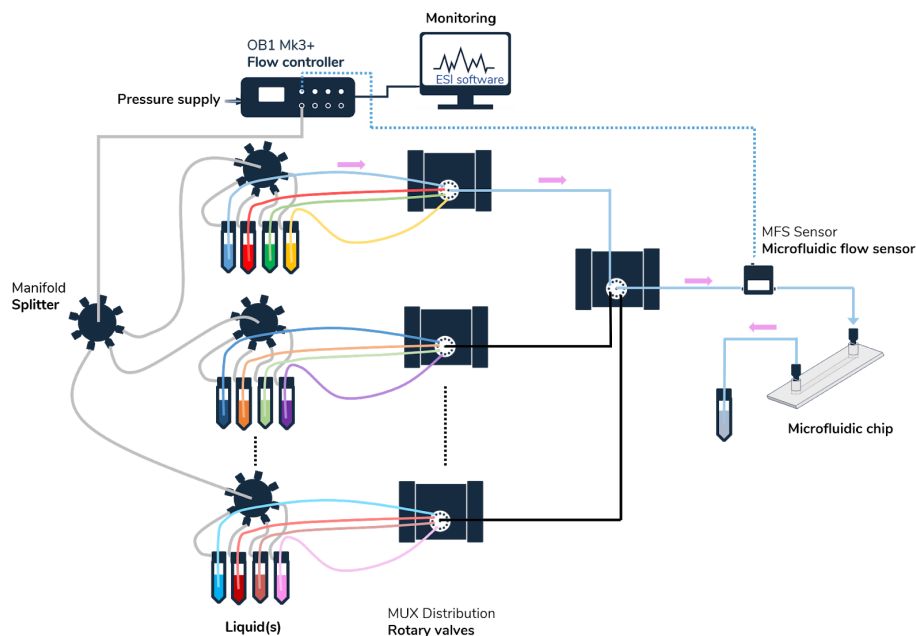
From 24 to 144 solutions

If the objective is to scale up your experiment significantly, it is possible to work with up to nearly 150 solutions by using 2 stages of Distribution valves and connect the outlets of the first set to the inlets of the second, as illustrated in the schematic below.

It is worth noting that up to 16 instruments can be connected to the ESI software. It is then possible to connect 15 MUX Distributions at the same time and an OB1 pressure controller.

TIP

If you add a line of pressure to the manifold, the pressure in the reservoirs will remain unchanged.



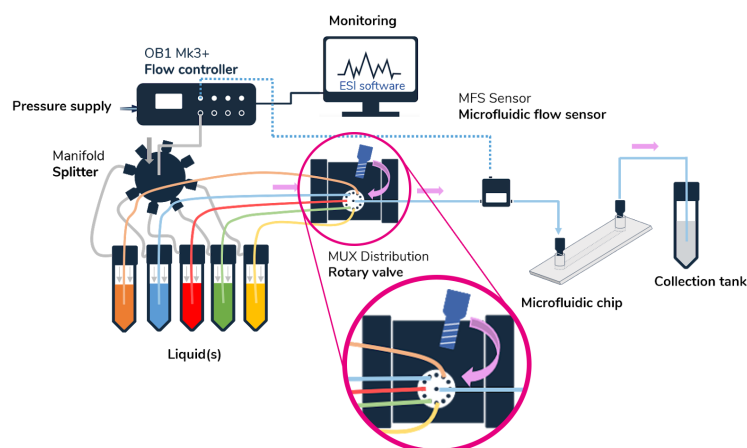
TIP

In order to ensure comparable flow rate conditions in the microfluidic device for all injected solutions at a given pressure, we recommend using the same length of tubing for all the microfluidic lines. To ensure a good control of the flow rate, the best solution remains the feedback loop with a sensor connected to the pressure controller (refer to appendix 2 for more information).

Appendix 6 Stop the flow in the device

The valve can also be used to accurately and almost instantaneously stop the flow in the device, e.g. for imaging. When the flow needs to be stopped in the device, rotate the MUX Distribution valve to a blocked port (using the plug fitting provided with the valves, such as port 12 in the example).

The flow will stabilize to zero faster than when applying a zero pressure or flow rate.

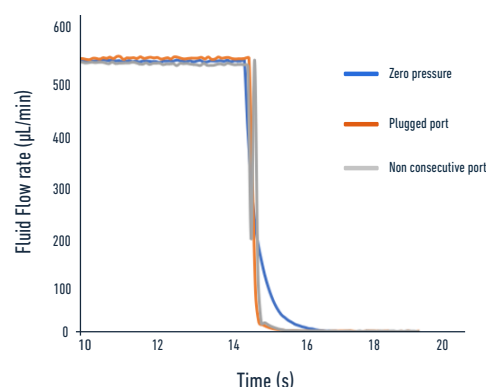


Regulator mode

When the valve rotates from port 1 to a port blocked by a plug fitting, using the mode "shortest" of the MUX Distribution, the flow rate stabilizes faster at zero ("Plugged port" curve) compared to the experiment where the applied pressure is set to zero ("Zero pressure" curve).

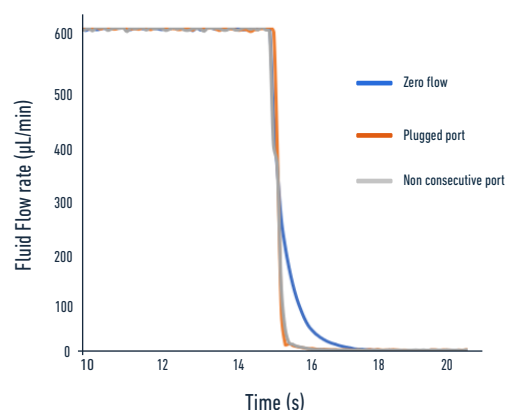
In the automated sequence below, step 5 was used to select a blocked port (valve 12). The response time is shown as "Plugged port" in the accompanying graph. The result of replacing this step by an OB1 command to apply zero pressure is shown as the trace "Zero pressure".

Switching between two non consecutive ports (one open and one blocked) does not impact the reactivity of the flow ("Non consecutive port" curve).



Sensor mode

Stopping the flow by rotating the valve to a blocked port ("plugged port" curve) is more efficient compared to applying a zero flow condition ("zero flow" curve).



If you are controlling the flow with the sensor mode, be aware that in this mode, the OB1 pressure controller regulates the pressure to keep the flow rate stable at a particular value. When the rotary valve is positioned on the blocked port, the flow will stop and the flow rate will be close to zero. However, the pressure on the OB1 will keep increasing in order to counterbalance this situation and try to keep the flow rate at the targeted value.

Appendix 7 Optimization of your sequential injection system using valves

In its product range, in addition to the MUX Distribution rotary valve, Elveflow also offers **2/2 and 3/2 valves**, controlled with a MUX Wire valve controller, as solutions for switching the flow.

Refer to **appendix 1 “Microfluidic connector guide”** to learn how to connect the valves and the MUX Wire

The valves can be added to the sequential injection system in order to add functionalities and fulfill some experimental requirements. The 2/2 valve has 2 ports and allows the switching of flow on and off. The 3/2 valve has 3 ports and enables the direction of fluid to or from different microfluidic lines.

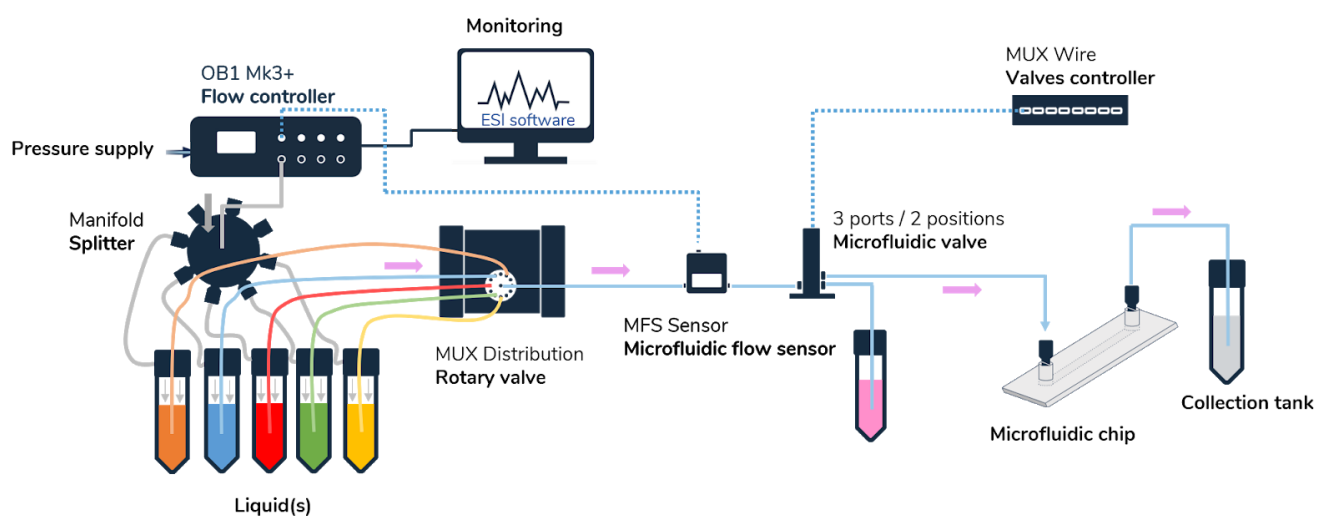
In both cases, the liquid can pass through the valves in one direction or the other without.



Avoid large diffusion effects in the tubing

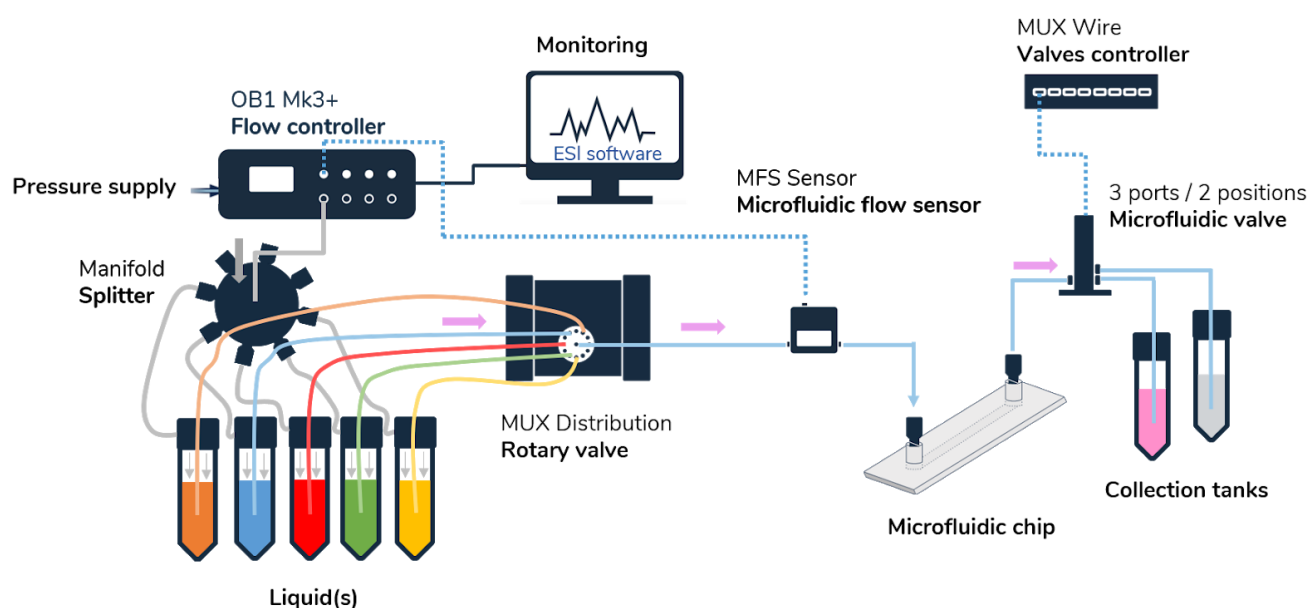
In order to decrease the effect of diffusion in the tubing, we recommend adding a 3/2 valve between the flow sensor and the microfluidic device (see the schematic). With this valve, a rapid flush is possible and hence the change of solution in your tubing will be quicker.

This configuration is also interesting in order to quickly flush air bubbles out of the microfluidic line.

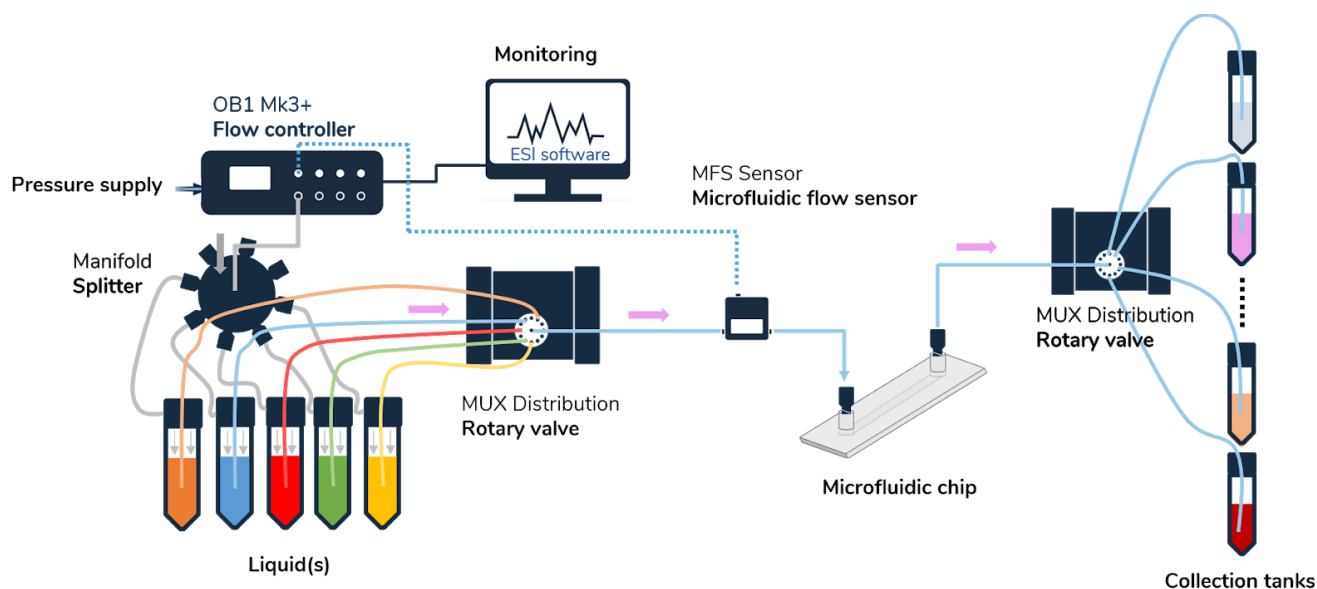


Flushing the device/collecting the sample in different outlets

For some experiments, it can be useful to flush the device between each injection, e.g. to clean the device or avoid cross contamination. By adding a 3/2 valve (3 ports, 2 ways) controlled by a MUX Wire at the outlet of the device, you can direct the fluid coming from the device in two different directions (e.g. a sample collection reservoir or a waste reservoir) or collect your samples in two different reservoirs. With the addition of this valve, it is possible to flush a solvent connected to the MUX Distribution through the device, then to rinse the entire microfluidic line and direct the solution to the waste.

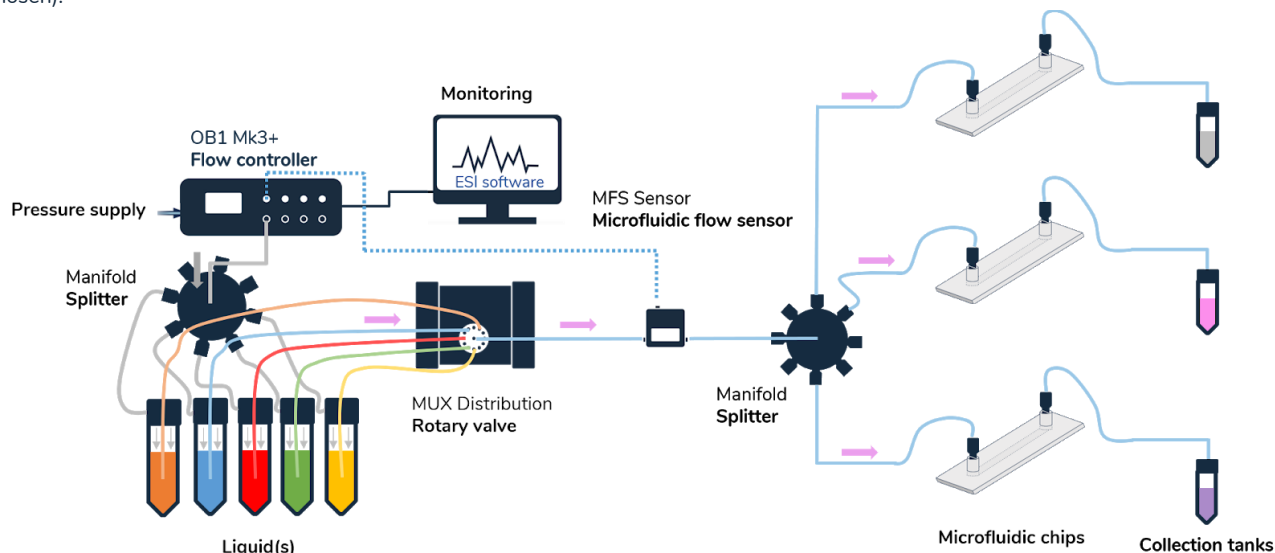


You can also use a second MUX Distribution valve at the outlet of the system in order to collect the product into 12 different reservoirs.



Parallelization

By adding a manifold at the outlet of the flow sensor, it is possible to increase the number of devices in which the solutions from the MUX Distribution are injected. It is then possible to parallelize numerous devices (the number depending on the type and number of manifolds chosen).

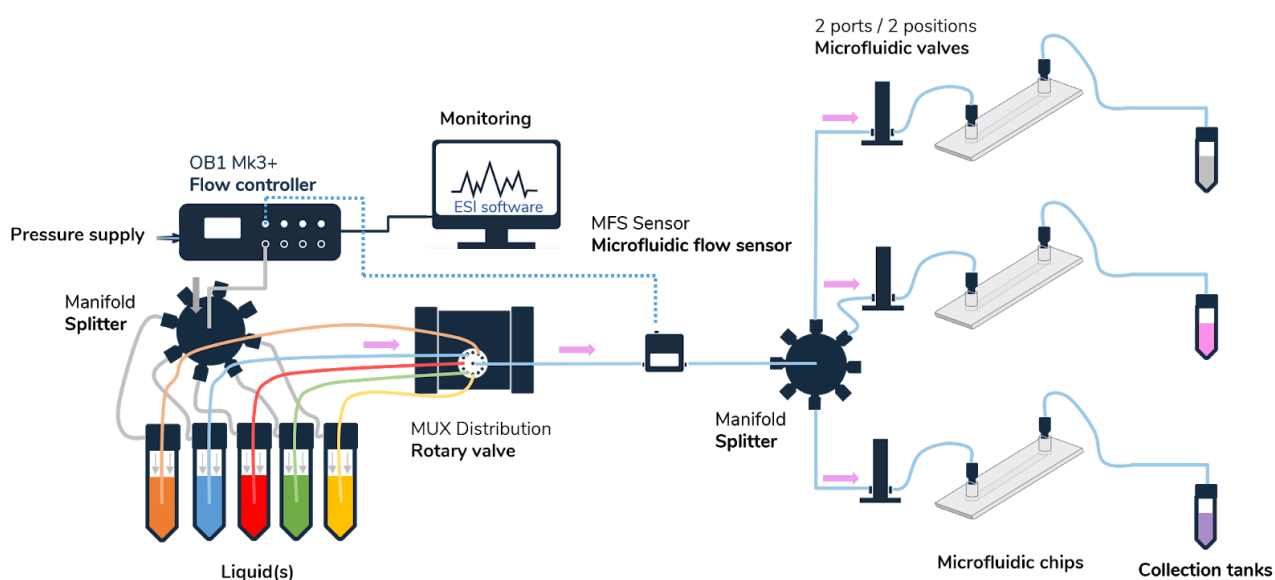


TIP

Be aware that the manifold is a passive system that doesn't ensure a similar flow rate in all the microfluidic devices. In order to ensure similar flow rate conditions, the fluidic resistance of the microfluidic lines coming from the manifold have to be similar. Hence, we advise you to use the same type and length of tubing.

TIP

Introduce 2/2 valves between the manifold and the devices to control which chip the solution is directed to by opening or closing the valves.



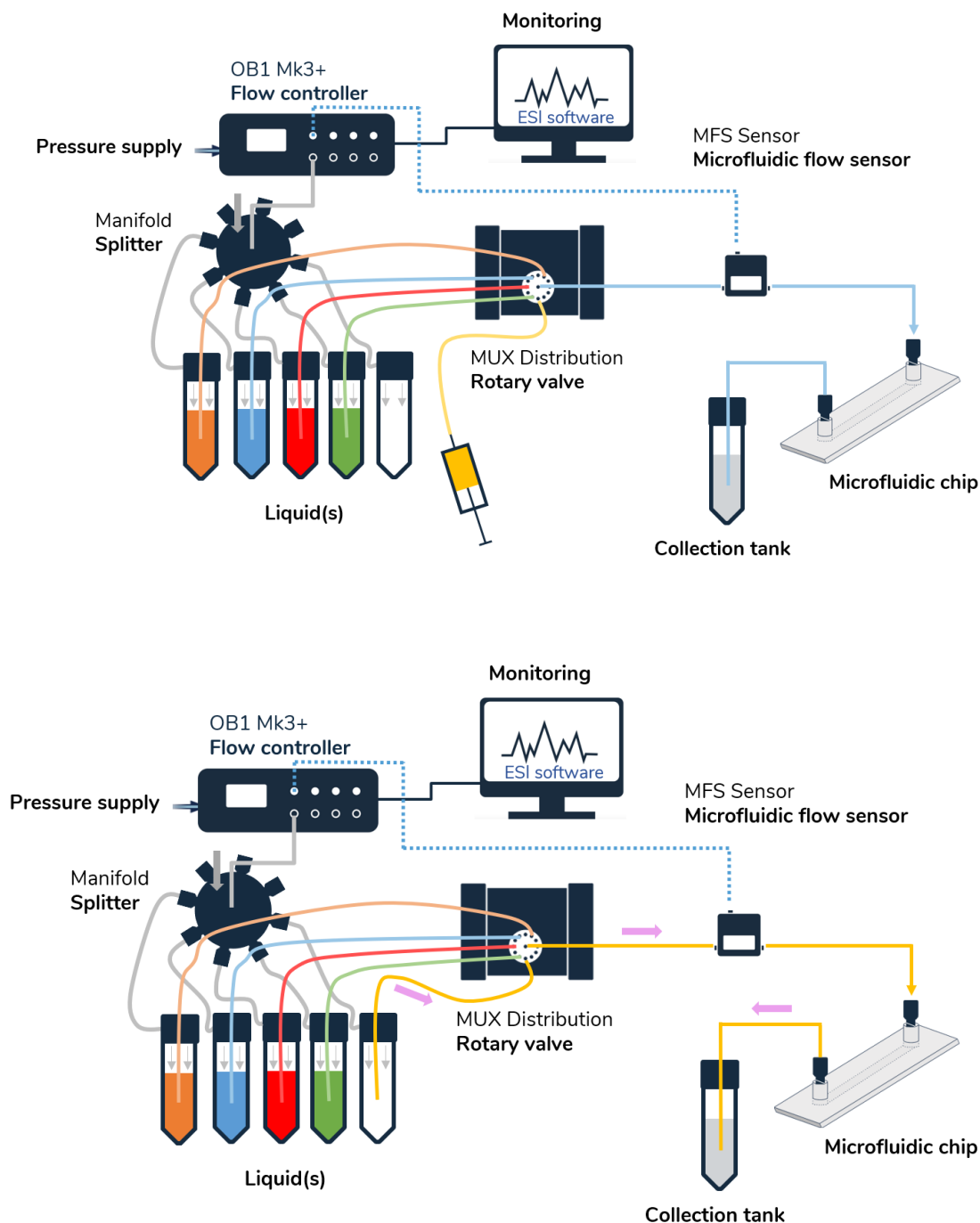
TIP

Parallelization of microfluidic devices can also be achieved by using the MUX Distribution to inject one liquid sample into 12 different microfluidic lines.

Appendix 8 Work with tiny volumes

If you plan to inject tiny volumes (10's of μL) in the microfluidic devices, consider preloading the tubing with a syringe and then using a pressurized reservoir filled with a buffer solution or no solution at all to push the targeted volume in the device.

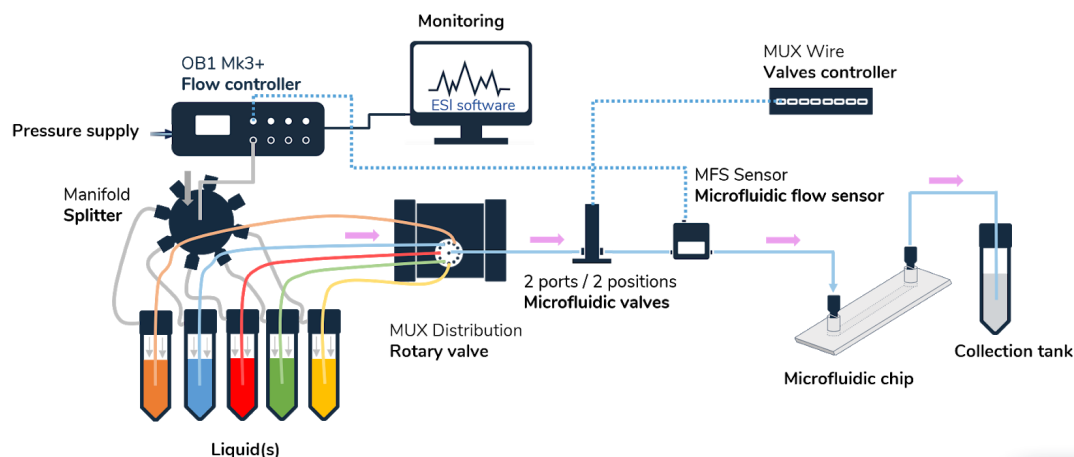
This method avoids the need for excess reagent simply to fill the reservoir.



Appendix 9 Lower the impact of the mechanical switch

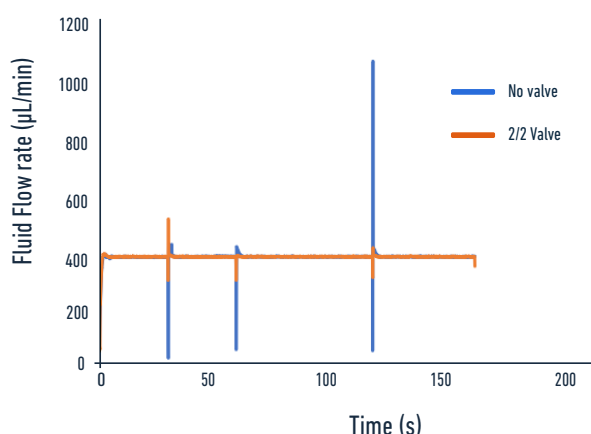
If the overshoot of the flow rate resulting from the mechanical switch of the rotary valve is impacting the accuracy of your experiment and you want to get rid of this effect, we advise you to add a 2/2 valve (on/off valve) between the MUX Distribution and the flow sensor. By closing the 2/2 valve

before applying the rotation of the MUX Distribution valve, and opening it again once the rotation is completed, the flow rate will be impacted less by the switch.

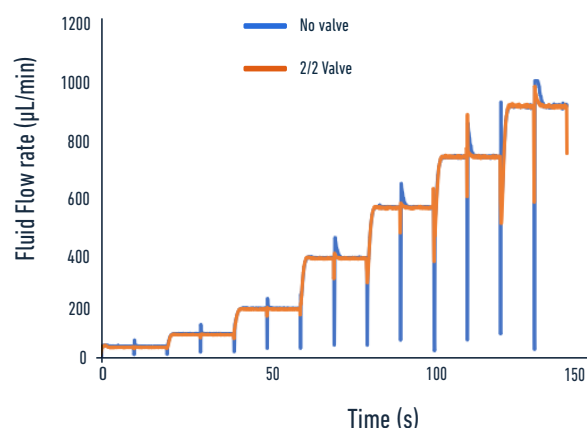


Below shows a comparison test using a simple automated sequence with and without the use of valves. The flow rate was fixed at 400 $\mu\text{L}/\text{min}$ and the Distribution valve was rotated from port 1 to port 2 and then to port 3. The 2/2 valve was closed right before the rotation and opened again right after. The table below shows the mean switching time and range of flow rate for the overshoot. The overshoot in flow rate decreased significantly (77% reduction) when a 2/2 valve was added to the setup, demonstrating its utility in this setup.

	Switching time (s)	Range ($\mu\text{L}/\text{min}$)
No valve	$2,33 \pm 0,41$	$594,9 \pm 297,6$
2/2 valve	$1,33 \pm 0,41$	$135,6 \pm 55,4$



A similar improvement can be obtained using a more complex automated sequence. In this example, the rotary valve was used to select a different liquid every 10s and flow rate was increased every 20s over the range 50 $\mu\text{L}/\text{min}$ to 1000 $\mu\text{L}/\text{min}$. Addition of the 2/2 valve significantly decreased the impact of the mechanical switch of the rotary valve on the flow rate.



```

1 Start Graph
2 OB1 Seq_Inj - Load: "OB1Seq_Inj_1..."
3 MUX Wire_Se... - Load: "MUXWire"
4 DIST Distrib... - Set valve: 1
5 MUX Wire_Se... - Load: "MUXWire"
6 30 s
7 MUX Wire_Se... - Load: "MUXWire"
8 DIST Distrib... - Set valve: 2
9 MUX Wire_Se... - Load: "MUXWire"
10 30 s
11 MUX Wire_Se... - Load: "MUXWire"
12 DIST Distrib... - Set valve: 3
13 MUX Wire_Se... - Load: "MUXWire"
14 1 min
15 MUX Wire_Se... - Load: "MUXWire"
16 DIST Distrib... - Set valve: 4
17 MUX Wire_Se... - Load: "MUXWire"
18 45 s
19 Stop Graph
20 OB1 Seq_Inj - Load: "OB1"
21 END End
  
```


Appendix 10

Automated control of the injected volume using the “flow integration” module

The aim of this section is to detail the use of the “**flow integration**” module in an **automated sequence** in order to directly control the injected volumes.

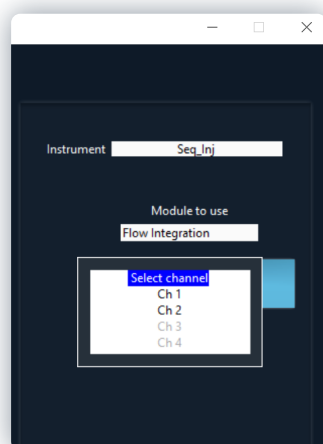
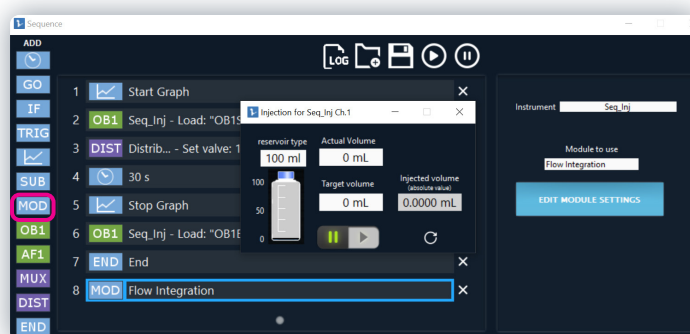
Before reading this appendix, we advise you to refer to **appendix 4** in order to learn the basics of automation with the ESI sequencer.

To create a sequence, click on the top middle button “Create Sequence” of the main window of the ESI software: the sequence window appears.

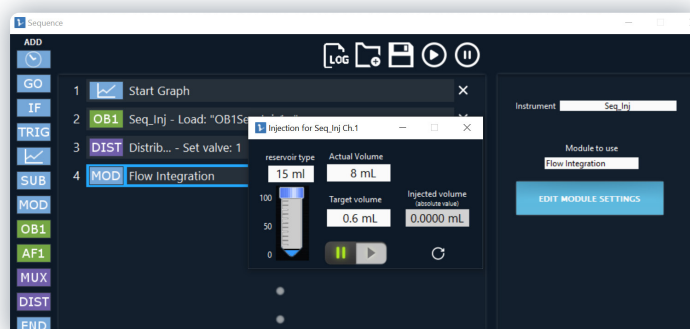
1 Create a step to record the data of your experiment with the Graph box (optional). Create a step for the OB1 (control in pressure or flow rate) and a step for the MUX Distribution in order to select the port corresponding to the solution to inject as described in appendix 4.

2 On the sequence window: on the left side of the window, click on the blue “MOD” box (a line “MOD” appears), on the right side, **select the instrument** (your OB1 pressure controller should appear when clicking on “Instrument”) and **choose the right module**, here “Flow integration”.

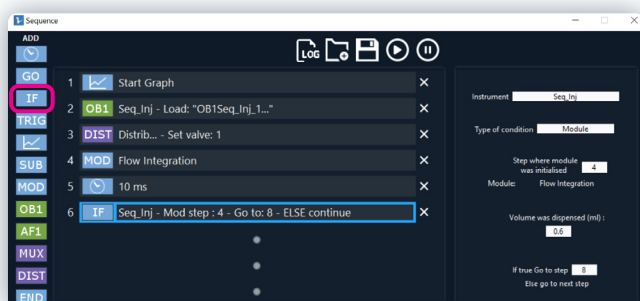
Click on the “Edit module settings” button. A new window will appear. Select the reservoir type used, indicate the actual volume of solution in the reservoir and the targeted volume. **Don’t click on the play button as it will be automatically triggered in the sequence.**



If several sensors are connected to the OB1, an additional window will appear when clicking on the “Edit module settings” button in order to **select the channel** to which you want to apply the module.



3 On the left side of the sequence window, click on the “Wait” box (clock symbol) and a new line with a time frame appears: **set a duration of 10 ms.**



4 On the left side of the sequence window, **click on the blue “IF” box**. A new step will appear. The goal of this step is to check if the volume to be dispensed has been reached or not.

On the right side, **select the instrument** (your OB1 pressure controller should appear when clicking on “Instrument”). **Choose the type of condition** (“Module” in this case), indicate the step number where the module was initialised (step 4 in the example) and the targeted corresponding volume of the initialisation step. Indicate also the next step to go in the sequence when the targeted volume is reached (“If true go to step 8 in this example”). At this point, if the targeted volume is not reached yet, the sequence will go to the step right after.

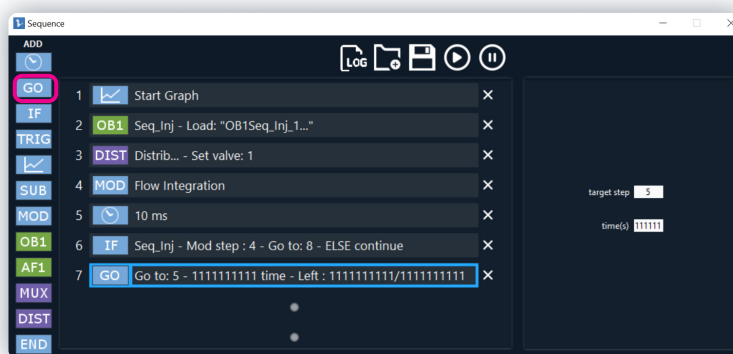
TIP

If you are using multiple modules or if you change the position of the steps during the creation of the sequence, be aware that the information regarding the targeted steps are not automatically updated. Always check one last time that the numbers and the targeted steps match your requirements before starting the sequence.

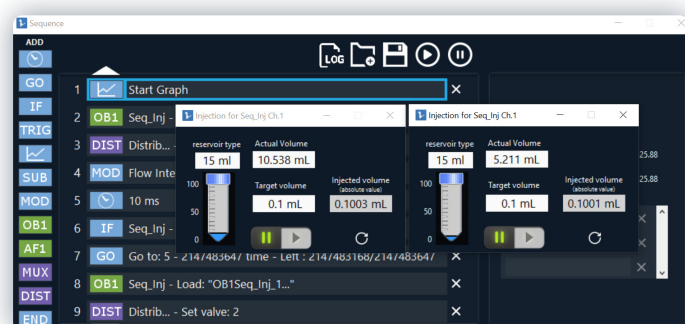
5

On the left side of the sequence window, click on the blue “GO” box. A new step will appear. This step allows you to create a loop in the sequence. As long as the volume is not reached, the sequence will stay in the loop; once the volume is reached, it will get out of the loop and keep going (this condition on the volume is checked in the “IF” step).

Choose and indicate the number of the “target step” (here, the wait step 5, in order to create a loop on the module command and keep injecting volume).



Indicate also in the “time” area how many times you want to repeat the loop. We recommend using a very high number to ensure that the targeted volume is reached before the loop is exited.

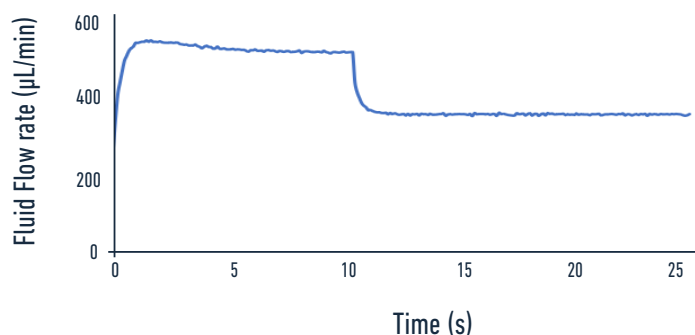


6

Repeat the successive steps “OB1”, “DIST”, “MOD”, “Wait”, “IF” and “GO” as many times as you like for all the solutions you want to inject.

In this example, we inject 0.1 mL of the solution connected to port 1 at 700 mbar and then 0.1 mL of the solution connected to port 2 at 500 mbar.

The flow is automatically maintained at 0 at the end of the sequence once the right volume has been injected.



**CONGRATULATIONS, YOU HAVE
NOW MASTERED THE AUTOMATED
INJECTION OF PRECISE VOLUMES!**

TIP

Once the targeted volume is injected with the module “Flow integration”, the channel is configured to maintain the flow at 0. Hence, if you want to inject another solution, you need to add a new line of command for the OB1 after the loop for the module “Flow integration”, even if the configuration of the OB1 remains the same.



Before launching the sequence again, you need to close all the “Flow integration” module windows and reinitialize the “MOD” step. Go to the button “Edit module settings” and press the reinitialisation button. The injected volume will be set again to 0.

Repeat the same operation for all the “MOD” steps. Otherwise, the sequence will not run correctly and the solution will not be injected as the injected volume will already be equal to the targeted volume.

Supplementary information

Conditions of use

Terms and conditions of use

We strongly believe in the intrinsic quality of our microfluidic instruments line and we hope that you will be pleased with your purchase. However, in the unlikely event that you should receive damaged or incorrect goods in your delivery, please notify us within 7 days.

You will be offered the option of a refund or an exchange (provided the goods are in stock).

You may be asked to return goods for inspection. In this case we will refund the shipping fees.

Should the damaged or incorrect item be no longer available, you will be given the option of a refund. Please note that goods that become damaged or broken after 7 days of receipt cannot be returned.

Unwanted items

If for any reason you do not wish to keep your purchase and would like a store credit, then please notify us within 7 days.

We cannot accept unwanted returns that have been opened, used or damaged by the customer.

For unwanted goods, we allow up to 14 days for the return of goods. We will only issue a credit

upon receipt of all returned goods.

Please note that we are unable to refund your costs in returning unwanted goods or the delivery costs of sending the goods to you in the first place.

Cancellations

If you wish to cancel your order please email us immediately: contact@elveflow.com.

Privacy Policy

Customer details remain private and confidential and will not be released to a third party unless required to do so by law.

We use the information we collect about you to process orders, to provide a more personalized shopping experience and, if you request it, to notify you about new products, special offers or other information that may be of interest to you. We do not sell or pass on any personal information to any other Companies or Organizations.

Payments & Procedures

Payment from private customers must be paid upfront. Trade orders from registered companies or organizations can be invoiced. Payment is

due strictly within 30 days of the invoice date.

Products & Prices

Please note that some goods may vary in style, color or detail from the image shown. We reserve the right to change prices at any time.

Transport and storage

Be careful not to harm or shake Elveflow® products while moving. Elveflow® products must not be transported when plugged in. Store products in standard conditions in an adapted box (typically the one used to send you the product).

Humidity and temperature must not exceed those of the specifications.

Exclusive remedies

The remedies provided herein are the customer's sole and exclusive remedies. Elveflow® shall not be liable for any direct, indirect, special, incidental, or consequential damages, whether based on contract, tort, or any other legal theory.

Safety Information

THE FOLLOWING GENERAL SAFETY PRECAUTIONS MUST BE FOLLOWED DURING ALL PHASES OF OPERATION, SERVICE, AND REPAIR OF THIS INSTRUMENT. FAILURE TO COMPLY WITH THESE PRECAUTIONS OR WITH SPECIFIC WARNINGS ELSEWHERE IN THIS MANUAL VIOLATES SAFETY STANDARDS OF DESIGN, MANUFACTURE, AND INTENDED USE OF THE INSTRUMENT. ELVESYS ASSUMES NO LIABILITY FOR THE CUSTOMER'S FAILURE TO COMPLY WITH THESE REQUIREMENTS.

Important advice

Elveflow® products are for research use only.

No liquid should get into the OB1, otherwise this would void the warranty.

The pressure source connected to the OB1 must be dry, dust and oil free, and of a maximum of 10 bar. Please take the required action to ensure that these conditions are met and maintained.

Conditions of use

This instrument is intended for indoor use. It is designed to operate at a maximum relative humidity of 60% and at altitudes of up to 2000 meters. Operating temperature range is +5 °C to 50 °C.

Do not operate in wet/damp conditions: to avoid electric shock, do not operate this product in wet or damp conditions.

Do not operate in an explosive environment. Do not operate the equipment in the presence of explosive or flammable gases or fumes.

Warning: Do not use this product as safety or emergency stop devices or in any other application where failure of the product could result in personal injury. The protective features of this product may be impaired if it is used in a manner not specified in the operating instructions. Before installing, handling, using or servicing this product, please consult the data sheet and user manual.

Failure to comply with these instructions could result in death or serious injury. If the buyer purchases or uses Elveflow® products for any unintended or unauthorized application, the buyer shall defend, indemnify and hold harmless Elveflow® and its officers, employees, subsidiaries, affiliates and distributors against all claims, costs, damages and expenses, and reasonable attorney fees arising out of, directly or indirectly, any claim of personal injury or death associated with such unintended or unauthorized use, even if Elveflow® is allegedly negligent with respect to the design or the manufacture of the product.

Pressurized Equipment

Care must be taken when the Elveflow® pump is pressurized to ensure that the instrument is not damaged in any way.

Protection

Safety glasses and lab coats should be worn at all times when using an Elveflow® pressure pump due to the use of pressurized equipment. This is particularly important when hazardous liquids are used.

Electricity Advice

Use Elveflow® instruments with the provided power unit only. Maintenance should only be attempted by qualified Elveflow® personnel. Removal of the back panel may invalidate any warranty.

Before applying power: verify that the line voltage matches the product's input voltage requirements and that the correct fuse is installed. Use only the specified line cord for this product and make sure the line cord is certified for the country of use.

Fuses: only fuses with the required rated current, voltage, and specified type (normal blow, time delay, etc.) should be used. Do not use repaired fuses or short-circuited fuse holders. To do so could cause a shock or fire hazard.

Keep away from live circuits: operating personnel must not remove instrument covers. Component replacement and internal adjustments must be made by qualified service personnel. Do not replace components with a power cable connected. Under certain conditions, dangerous voltages may exist even with the power cable removed.

To avoid injuries, always disconnect power, discharge circuits and remove external voltage sources before touching components.

ESD precautions: the inherent design of this component causes it to be sensitive to electrostatic discharge (ESD). To prevent ESD-induced damage and/or degradation, take

customary and statutory ESD precautions when handling this product.

Maintenance advice

Maintenance should only be attempted by qualified Elveflow® personnel. Removal of the back panel will invalidate any warranty.

Do not service or adjust alone: do not attempt internal service or adjustment unless another person, capable of rendering first aid and resuscitation, is present.

Do not substitute parts or modify the instrument: because of the danger of introducing additional hazards, do not install substitute parts or perform any unauthorized modification to the instrument.

Return the instrument to an Elveflow® Technologies Sales and Service Office for service and repair to ensure that safety features are maintained.

Instruments which appear damaged or defective should be made inoperative and secured against unintended operation until they can be repaired by qualified Elveflow® personnel.

CE compliance

This product meets the essential requirements of applicable European Directives, as amended for CE marking, as follows:

Electromagnetic Compatibility

COUNCIL DIRECTIVE 89/336/EEC of 3 May 1989

This directive has been amended by the following Council Directives:

1. 92/59/EEC of 29 June 1992 (General Product Safety)
2. 93/68/EEC of 22 July 1993 (CE Marking directive)
3. 99/5/EC: Directive of Radio Equipment & Telecommunications Terminal Equipment (R&TTE).



Technical support

customer@elveflow.com

General information

contact@elveflow.com

+33(0).184.163.807

ELVESYS – Microfluidics innovation center

172 Rue de Charonne,
75011 Paris, FRANCE

www.elveflow.com

Product:

Elveflow.com/microfluidic-products/microfluidics-application-packs/sequential-fluid-injection-pack/



MAY THE FLOW BE WITH YOU