

Application Bulletin 363

Installation Instruction for ProfIC Vario 1 PCR-UV/VIS

The ProfIC Vario 1 PCR-UV/VIS is a Professional IC system for the fully automatic determination of UV/VIS active compounds after post-column reaction (PCR). «ProfIC Vario 1» safely masters all routine tasks in ion chromatography. It is simple to use and extremely reliable.



Table of contents

1.	Delivery Package	
	IC	2
	Detector	2
	Sample processor	2
	Optional	2
2.	Installation	2
2.1.	Installation of the software	
2.2.	Accessory Kit: Vario/Flex Basic (6.5000.000)	2
2.3.	Accessory Kit: Vario/Flex ONE (6.5000.010)	2
2.4.	858 Professional Sample Processor	2
2.5.	Interconnection of devices	3
2.6.	Professional IC Vario	3
3.	MagIC Net	4
3.1.	Configuration	4
3.2.	Method adjustment	4
3.3.	Purge of the system	4
3.4.	User defined determination	4
4.	Exemplary measurement	6
5.	Alternative PCR transportation	6
5.1.	High-pressure pump	6
5.2.	Dosino	6
6.	Optional equipment	7
6.1.	Liquid Handling Station:	7
6.2.	Eluent Production Module	8



1. Delivery Package

Delivered with ProfIC Vario 1 PCR-UV/VIS package:

Nr	Article no.	Article designation			
IC					
1	2.940.1110	Professional IC Vario ONE/Prep1			
1	2.943.0110	Professional Reactor Vario			
Detector					
1	2.944.0010	Professional UV/VIS Detector Vario			
Sample processor					
1	2.858.0020	Professional Sample Processor			
Optional					
1	6.6059.302	MagIC Net 3.0 Professional CD - 1 License			
1	6.6059.303	MagIC Net 3.0 Multi - 3 licenses			
1	6.2041.760	Sample rack 54 x 11 mL + 1 x 300 mL			
1	6.2041.440	Sample rack 148 x 11 mL + 3 x 300 mL			
1	6.2743.040	Sample tubes 2.5 mL			
1	6.2743.050	Sample tubes 11 mL			
1	6.2743.070	Stopper with perforation			
1	6.1020.230	Metrosep A Supp 10 - 250/2.0			
1	6.1020.600	Metrosep A Supp 10 Guard/2.0			
1	6.1825.250	PEEK sample loop 1 mL			
1	2.941.0010	Eluent Production Module			
1	6.2769.110	Sensor empty 2 L			
1	6.5330.130	Kit for LH station			

2. Installation

Following is a detailed description of how to install ProfIC Vario 1 PCR-UV/VIS.

We strongly recommend that the individual steps are carried out in the order given below

2.1. Installation of the software

All programs must be shut down first. Make sure no Metrohm instrument is connected to the PC. Install MagIC Net with the help of the MagIC Net - CD. The Microsoft Installation Wizard is accepted and executed. All the standard directories proposed by the program should be accepted. Restart windows.

Now as soon as you connect a new Metrohm instrument via USB connection to the PC, the driver is installed and a window will pop up in MagIC Net, asking you if you'd like to store this device in your configuration. If you do, please

answer with yes. The names will be checked later in this installation instruction.

2.2. Accessory Kit: Vario/Flex Basic (6.5000.000)

Using the Accessory Kit Vario/Flex Basic, install the Professional IC Vario. Remove handle, remove the bottle holder on top of the IC and place the reactor and the UV/VIS detector module. There is also the possibility to place both modules beneath the IC. For this, lay the IC on its side, remove the base plate, place the reactor and the detector underneath (make sure to have the reactor as an inbetween) and fix the base plate back underneath the tower. Now place it in an upright position again. In this application bulletin all steps will be described for the placement of the reactor and detector on top of the IC. Please refer to the manuals 944 Professional UV/VIS Detector Vario and 943 Professional Reactor Vario for more details.

After retightening the bottle holder on top of the detector, connect the detector and the reactor with the IC using the two USB cables (6.2151.020), remove the transport locking screws, connect the leak sensor cable and connect the drainage tubing.

In continuation, set up the waste collector by assembling the cap and screwing it onto the vessel. Then hang the waste collector holder on the side of the IC, so you can observe the droplets coming out of the capillaries later connected to the collector. Attach the waste tube to the vessel and lead it to the waste canister. When the tube is too long, please shorten it, because it is important to have a high level difference for the liquid to drain.

The power cables and USB cables (6.2151.020) are plugged into the rear of the Professional IC Vario, the 943 Professional Reactor Vario and the Professional UV/VIS detector Vario. Please don't switch on the instrument yet. This step will follow after the completed installation.

2.3. Accessory Kit: Vario/Flex ONE (6.5000.010)

In the box with the Accessory Kits ONE, you will find all the accessories for setting up the eluent bottle. Please lead the aspiration tube for the eluent through the M8 stopper, the earring and the eluent cap. Then fix the white weight (6.2744.210), the adaptor (6.2744.210) and the aspiration filter (6.2821.090) on the eluent aspiration tube, all the while being careful not to touch the filter and its connections with bare hands in order to avoid cross contamination. Also fix the filled adsorber tube on the eluent cap. Please refer to the 940 Professional IC Vario manual for a stepwise description.

2.4. 858 Professional Sample Processor

For a detailed description, please refer to the 858 Professional Sample Processor manual. In general, you will



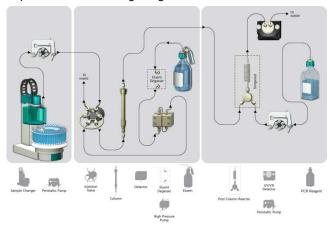
have to do the following: Plug in the Swing Head connection cable and the power supply cable. The controller cable (6.2151.000) is plugged into the plug "Contr." on the 858 and connected to the IC device via USB. The sample processor initializes and lifts its Swing Head, once the IC device it is connected to is recognized by the software. For this, you have to plug the USB cable of the IC into the PC and turn it on. Afterwards, it is possible to mount the retaining plate, the needle and the safety shield (for detailed instructions please refer to the 858 Professional Sample Processor manual).

For the following installations, please shut down the IC again and disconnect the 858 from the power supply.

Subsequently, insert the pump tubing (white/white 6.1826.360) into the peristaltic pump and connect the aspiration side to the needle via a PEEK capillary (6.1831.160) (see also 858 manual).

2.5. Interconnection of devices

The whole setup of the ProfIC Vario 1 PCR - UV/VIS is depicted in the following image:



Prepare and degas an eluent suitable for the column you want to use (see column manual). For example, chromate can be separated on a Metrosep A Supp 10 - 250/2.0 with an eluent of ammonium sulfate (250 mmol/L) and ammonium hydroxide (100 mmol/L).

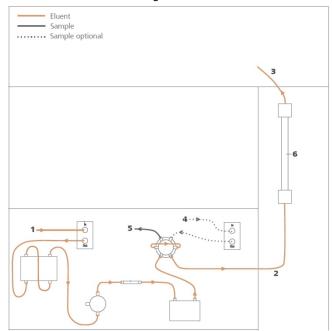
Prepare an adequate PCR solution. For example, chromate can react with a solution of 1,5-diphenylcarbazide (2 mmol/L), methanol (10%v/v), and sulfuric acid (2.7%v/v).

2.6. Professional IC Vario

Capillaries are connected according to the following list and the diagram below (also see manual):

- 1. Connection to the Eluent bottle
- 2. Capillary to column inlet 0.25 mm ID
- Column outlet capillary 0.25 mm ID, to be connected to the Y-connector of the reactor

- 4. Sample aspiration capillary PTFE, 0.5 mm ID. The sample degasser is only used if necessary
- Sample outlet capillary PTFE, 0.5 mm ID, ca.
 1 m, connected to waste container
- The UNF 10/32 coupling (6.2744.040) is installed instead of the column to rinse the system with eluent. After rinsing the column is installed.



In order to transfer the sample to the IC device, place a PTFE capillary (6.1803.030) of suitable length (~1 m) between the peristaltic pump tubing outlet of the sample processor and the valve of the IC instrument. The sample injection is at position 1 of the valve (see diagram of chapter 2.1, capillary number 5). A counter current flow of the sample with regard to the eluent flow is recommended, in order to minimize diffusion and carry over.

The column outlet is connected with the Y-connector of the reactor. The PCR solution is transported with a peristaltic pump. Thus, install a peristaltic pump tubing (orange-green 6.1826.310) and connect its outlet-side to the second opening of the Y-connector of the reactor.

The reactor outlet is connected with the detector "in". The detector outlet capillary has to be at least 1 m long (0.25 mm ID) to guarantee a stable pressure in the flow-through cell.

Make sure that all outlet capillaries are put into the waste collector (detector outlet, sample waste).



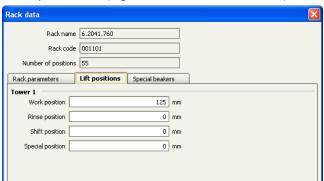
3. MagIC Net

3.1. Configuration

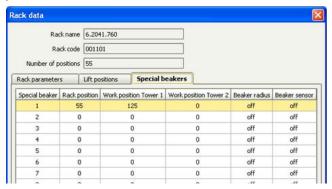
Now please connect the USB and power cables from the instruments and turn their power on. Connected USB devices are automatically recognized when MagIC Net is started. After confirmation of the automatically generated requests, the devices and columns are stored in the configuration. The devices are predefined as "940 Professional IC Vario 1", "943 Professional Reactor Vario 1", "944 Professional UV/VIS Detector Vario 1", and "858 Professional Sample Processor 1". Name them accordingly, if other names appear in your configuration (e.g. due to changed setting on your computer).

In the window configuration, the 940 Professional IC Vario, the 943 Professional Reactor Vario, the 944 Professional UV/VIS Detector Vario, the 858 Professional Sample Processor and the column Metrosep A Supp 10 - 250/2.0 should now be visible. Add and define eluent and PCR solution.

The settings for the rack require a "work position" fitting to the respective rack (e.g. 125 mm for rack 6.2041.760).



Also define a special beaker for rinsing the needle. Fill the beaker with ultra-pure water and place it at the defined position on the rack.



3.2. Method adjustment

In the window method, import the method for ProfIC Vario 1 PCR-UV/VIS from the installation CD: Go to File \rightarrow method manager and choose your method group. Afterwards, click on edit \rightarrow import and choose the following pathway on the

installation CD:

MagICnet\examples\methods\ProfIC Vario\ProfIC Vario_1_ PCR_UVVIS. imet.

In the window method, open the method ProfIC Vario_1_PCR_UVVIS and assign the following equipment: Assign the column to the analysis UVVIS, define the rack of the connected sample processor, and assign the eluent to the high-pressure pump and the PCR solution to the peristaltic pump.

In the example method the wavelength and bandwidth are predefined for the chromate analysis, to change those, chose your UV/VIS detector in your method. Under "Detector" you'll be able to configure up to 8 parallel channels with their respective wavelengths and bandwidths. Additionally one reference channel can be defined.



Perform a method test and save the method.

3.3. Purge of the system

Before inserting the column, flush the system for about 10 minutes and get rid of air bubbles (by using the purge valve and syringe).

Now insert and rinse the precolumn for 10 minutes by leading the outlet directly into the waste. Afterwards connect the analytical column and flush it likewise for 10 minutes. Only afterwards the column is completely connected to the flow path of the eluent (see also 940 manual).

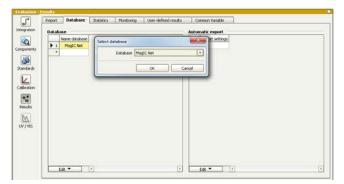
In order to start the equilibration, go to the window work place, load the method, and press "Start HW".

Equilibrate the system until the baseline is stable.

3.4. User defined determination

In the window method, enter the ions and the concentrations of the required standards. Now please add the correct Database in the Evaluation window (Evaluation - Results - Database) for the method.





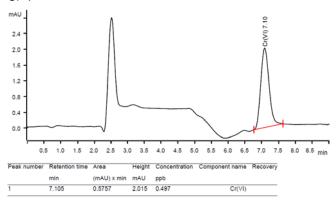
In the window work place, equilibrate the system until the baseline is stable. Set up a "determination series", describing your samples by ident, vial number, sample type (standard, blank, or sample etc.). After putting the analyte solutions onto the rack, press "start".

For evaluation and after recording the first chromatogram, check the retention times of your compounds. Since they depend on the performance of your column, you may have to adjust them in your method.

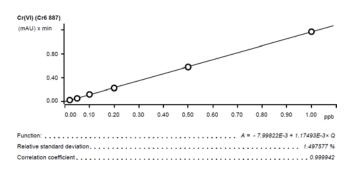


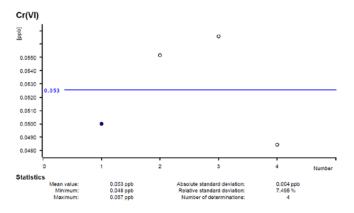
4. Exemplary measurement

Chromate (Cr^{VI}) was analyzed in low concentrations. The following chromatogram shows a 1 mL injection of 0.5 μ g/L Cr^{VI} .



The calibration is linear over a wide calibration range and typically has correlation coefficients of about 0.9999 and standard deviations of about 1.5%. Low concentrations such as 0.05 μ g/L can be reproducibly determined (e.g. see figures below).





5. Alternative PCR transportation

In the previous example, the PCR solution is transported with a peristaltic pump. Alternatively, it can also be transported with a Dosino or a high-pressure pump.

5.1. High-pressure pump

The high-pressure pump can be installed as extension module (e.g. 2.942.0040 Extension Module Vario HPG) or as plug-in unit of the chromatograph (e.g. 6.5865.040 Upgrade IC Vario HPG). For optimal operation of the high-pressure pump's pulsation dampener, a minimal backpressure of 2 MPa is required. This can be obtained by installing one or two back-pressure columns (6.1015.100 Metrosep BP 1 Guard/2.0).

A high-pressure pump guarantees the most stable baseline and shows the lowest noise. The flow is easily adjusted and controlled and has a high long-term stability (whereas the peristaltic pump needs careful flow adjustments and the tubing wears out after some time). However, when working with aggressive PCR solutions, it is recommended to rinse the high-pressure pump before shutting down the instrument.

5.2. Dosino

Furthermore, it is possible to transport the PCR solution with a Dosino. Baseline noise and stability are comparable to the setup with a peristaltic pump. The Dosino and the flow path can be automatically rinsed after a sample series. To do so, a rinse solution can be attached to the Dosino's port 3 and a waste bottle can be attached to its port 4 (presumed that the PCR solution is on port 2 and the capillary to the reactor is on port 1).

However, the Dosino refills before each chromatogram, which implies stopping of the PCR flow. Thus, there is a delay until pressure and baseline re-stabilizes. Normally, this is done during sample preparation, so it is not recorded in the chromatograms. Choose the dosing unit as small as possible (e.g. 2 mL for 0.2 mL/min typically), because a smaller step size leads to a smoother baseline. Nevertheless, the volume of the dosing unit must be high enough to provide PCR solution throughout the chromatogram.

Dosinos are sensible against pressure. So, any unnecessary back-pressure must be excluded to avoid leakage at the Dosino.



6. Optional equipment

6.1. Liquid Handling Station:

The Liquid Handling Station (LQH Station) consists of two function units: The rinsing and the dilution unit. As an addon it is mainly useful for rinsing the needle on the inside and outside, thus minimizing contamination.

The following kit is needed:

Nr	Article no.	Article designation
1	6.5330.130	IC Equipment: LQH Station



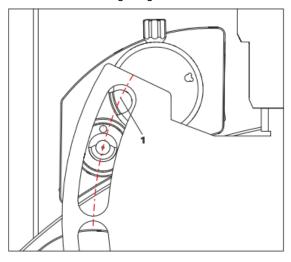
The kit includes:

Nr	Article no.	Article designation
1	6.1014.200	Metrosep I Trap 1 - 100/4.0
1	6.1602.160	Eluent bottle cap GL 45
1	6.1608.070	Eluent bottle /2 L / GL 45
1	6.1619.000	Adsorber tube for Dosing Unit
1	6.1624.000	Adapter SGJ 14
1	6.1826.390	Pump tubing LFL (yellow/yellow) 3 stoppers
1	6.1831.180	PEEK capillary i.D. 0.5 mm, 3 m
1	6.2744.010	Pressure screw 5x
1	6.2744.034	Coupling nozzle UNF 10/32
1	6.2841.120	Liquid Handling Station left

The installation of the LQH Station is done in two steps: First mount and align the Liquid Handling Station on the left hand side of the Sample Processor. To accomplish this, remove the sample rack and place the Liquid Handling Station on the black rail of the Sample Processor. Secure it temporarily in place with the screw and then loosen the small screws of the foot. The rack is replaced on the Sample

Processor and the small screws are fixed in a way that the approximate distance between LHS and rack is 0.5-1 mm.

Now the Liquid Handling Station needs to be aligned with the retaining plate. For this, loosen the big screw again and move the Station underneath the retaining plate. Looking from above you should now be able to see the small hole of the rinsing unit and part of the big hole of the dilution vessel, similar to the following image.

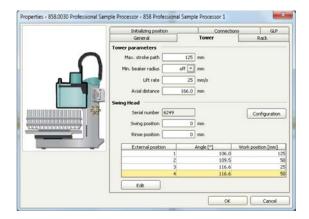


When you have found the correct position, please tighten the screw to fix the LQH Station completely to the Sample Processor.

In a second step, the work positions for the sample tube need to be defined in the configuration.

Please go to the configuration of the Sample Processor. Under Tower it is possible to define external positions of the swing head. For rinsing purposes, only the External Position 1 (small inner tube) and the External Position 2 (outer tube of rinsing unit) have to be defined. The proposed angles are only guiding values; they need to be adapted for every system separately. This can be done easily by adjusting the angles in the manual control window. As soon as you have found the correct angles, make sure to save them in the configuration. The work positions are fixed for all of the different setups.





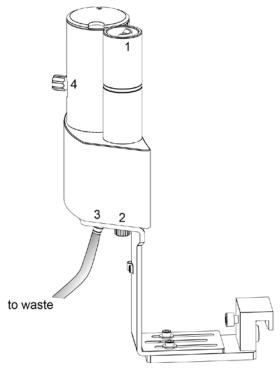
Depending on the available parts, the water supply to the Liquid Handling Station is managed differently. The inner tube of the rinsing unit is meant to be filled constantly with fresh Ultrapure Water, so the aspiration needle can be thoroughly cleaned from the outside.

All possibilities have the waste tube in common, Number 3 in the following image. Please fix the PVC tubing (6.1801.120) on the disposal connector on the bottom of the Liquid Handling Station.

If there is a peristaltic pump canal free, there is the possibility to connect a water bottle with a peristaltic pump tubing and lead a capillary to the PEEK pressure screw on the bottom of the Liquid Handling Station (Number 2 on the following schemata). Between the peristaltic pump and the LQH Station, please install an I-Trap (6.1014.200) into the flow path to cleanse the water before it goes into the IC system.

Another possibility arises with a Dosino:

Similar to the setup with the peristaltic pump before, connect one of the Dosino ports with a capillary to the PEEK pressure screw (Nr. 2) on the Liquid Handling Station. Then use the Dosino to push water from below into the rinsing unit when needed. Here, it is recommended to install an I-Trap (6.1014.200) into the flow path of the water, in order that the water is cleansed from all impurities that could build in the water reservoir.



Number 4 of the picture above shows the attachment point of a capillary in case of a dilution feature and Number 1 indicates the rinsing entity.

In the time program you can treat the external positions like a special beaker: Just move to the wanted angle, go into work position and start the Dosino or the peristaltic pump. Make sure to always dispose the waste in the external position 2.

6.2. Eluent Production Module

The 941 Eluent Production Module creates fresh new eluent out of eluent concentrate and ultrapure water. For installation instructions and further information, please refer to the 941 Eluent Production Module Manual.