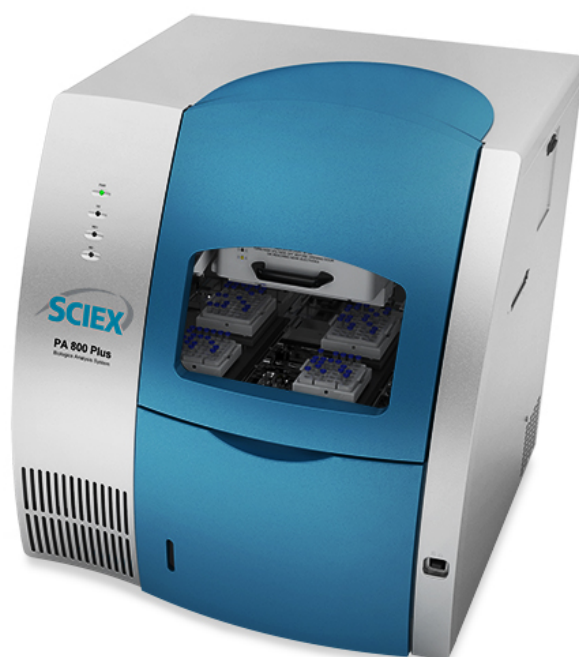


PA 800 Plus Empower™ Driver

User Guide



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This document gives instructions to use the Waters Empower™ software with a PA 800 Plus system. The PA 800 Plus Empower™ Driver must be installed on the computer with the Waters Empower™ software. For installation instructions, refer to the document: *PA 800 Plus Empower™ Driver Release Notes*.


This document includes instructions to calibrate the detectors in the PA 800 Plus system. Instructions for direct control of the PA 800 Plus system using the Waters Empower™ software are also given.

Note: For instructions about how to use the system safely, refer to the document: *Overview Guide*.

The Waters Empower™ software can also be used with the CESI 8000 Plus High Performance Separation-ESI Module when a laser-induced fluorescence (LIF), photodiode array (PDA), or ultraviolet (UV) detector is installed.

Related Documentation

This document is for users with some knowledge of the Waters Empower™ software. For information about the general features of the Waters Empower™ software, do this:

- Refer to the documentation supplied with the software.
- In the Empower Start dialog, click .
- In any of the Waters Empower™ software windows, click **Help**.

For instructions about how to use the Waters Empower™ software for a specified capillary electrophoresis (CE) application, refer to the following application guides:

- *Capillary Isoelectric Focusing (cIEF) Analysis Application Guide*
- *Fast Glycan Labeling and Analysis Kit Application Guide*
- *IgG Purity and Heterogeneity Assay Kit Analysis Application Guide*

For a general introduction to the PA 800 Plus system, refer to the document: *Overview Guide*.

For instructions about PA 800 Plus system maintenance, refer to the document: *Maintenance Guide*.

Waters Empower™ Software Terminology for 32 Karat Software Users

We recommend that users who have used the PA 800 Plus system with the 32 Karat software become familiar with the Waters Empower™ software terms.

Table 1-1 Waters Empower™ Software Terminology for 32 Karat Software Users

32 Karat Software Term	Waters Empower™ Software Equivalent	Description
No equivalents in 32 Karat software	Instrument Method	A method that contains the system parameters required for data acquisition. Parameters groups are general parameters, detector parameters, and a time program.
	Processing Method	A method that contains data processing parameters.
	Reporting Method	A method to create a report that shows the results of the processing method.
Method	Method Set	A combination of an instrument method, a processing method, and a report method. Processing and report methods are optional.
Sequence	Sample Set Method	A list of samples and related method sets that is sent to the PA 800 Plus system for data acquisition. Optionally, the Waters Empower™ software can do post-acquisition data processing and generate reports.
Report	Report	A file that contains information about the results of the data acquisition. Reports can also include information about the organization that generates the data. The layout and appearance of a report can be customized and saved as part of a report template.
Sample Tray	Plate	The tray or 96-well plate that holds the samples to be analyzed.
Buffer Tray	Plate	The tray that holds the vials that contain the buffer and rinse solutions.

**Table 1-1 Waters Empower™ Software Terminology for 32 Karat Software Users
(continued)**

32 Karat Software Term	Waters Empower™ Software Equivalent	Description
Controller	LAC/E module	The computer that controls the PA 800 Plus system.

PA 800 Plus Empower™ Driver License

To collect and analyze data with the PA 800 Plus Empower™ Driver, a USB license key is required. Install the USB license key in a USB port on the Waters Empower™ software LAC/E acquisition server.

If the license key is not present, then all of the controls in the Direct Control pane are disabled. Also, data acquisition will not start. If the license key is removed during data acquisition, then acquisition for the active method set completes but no additional data acquisition will start.

If required, then the license key can be removed from one LAC/E acquisition server and installed in a USB port on another computer.

Direct Control

2

This section shows how to use the Direct Control pane in the Waters Empower™ software to control the PA 800 Plus system.

The Direct Control pane has the following three sections, from top to bottom:

- **Instrument status pane:** Shows the status of the system. Refer to the section: [Instrument Status in the Direct Control Pane](#).
- **Status field:** Shows the status of the system or any process that occurs on the system. Error messages are also shown in red text in this field.
- **Parameter tabs and buttons:** Set the parameters for the system. Different tabs show for different detectors. Refer to the section: [Parameters and Buttons in the Direct Control Pane](#).

Figure 2-1 Direct Control Pane (PDA Detector)

Control Voltage 0.000 kV Current 0.000 µA Power 0 W	Temperature Cartridge 25.6 °C Storage 25.0 °C	Tray BI: A1 BO: - - Lamp Hrs D2: 7.00 h Hg: 0.06 h	PDA Detector <table border="1"><thead><tr><th>Chan</th><th>Wv</th><th>Bw</th><th>Absorb.</th></tr></thead><tbody><tr><td>Ch1</td><td>214</td><td>10</td><td>0.000000</td></tr><tr><td>Ch2</td><td>214</td><td>10</td><td>0.000000</td></tr><tr><td>Ch3</td><td>214</td><td>10</td><td>0.000000</td></tr><tr><td>Ref.</td><td>214</td><td>10</td><td>0.000000</td></tr></tbody></table>	Chan	Wv	Bw	Absorb.	Ch1	214	10	0.000000	Ch2	214	10	0.000000	Ch3	214	10	0.000000	Ref.	214	10	0.000000
Chan	Wv	Bw	Absorb.																				
Ch1	214	10	0.000000																				
Ch2	214	10	0.000000																				
Ch3	214	10	0.000000																				
Ref.	214	10	0.000000																				
Pressure 0.0 psi ➡ +	Coolant OK	Lamp On																					

Status Idle

Voltage Settings	Temperature	Lamp On/Off	Spectrum	Lamp Hour	Home
<div>Trays<ul style="list-style-type: none">Inlet Sample Tray 48 VialsInlet Buffer Tray 36 VialsOutlet Sample Tray 96 Positions/No TrayOutlet Buffer Tray 36 Vials</div>					Load
					Set
					Stop
					Advanced

PA 800 Plus

Instrument Status in the Direct Control Pane

Note: Pressure values can be shown in millibar (mbar) or pounds per square inch (psi). The registry setting for the Waters Empower™ software controls which value is used. The default unit is millibar. To change the units, refer to the document: *PA 800 Plus Empower™ Driver Release Notes*.

Figure 2-2 Instrument Status in the Direct Control Pane (LIF Detector)

Control		Temperature		Tray		LIF Detector	
Voltage	0.000 kV	Cartridge	24.8 °C	BI:	A1	Channels	RFU
Current	0.000 µA	Storage	25.0 °C	BO:	A1	Ch1	0.000000
Power	0.000 W			Laser Hrs		Ch2	0.000000
Pressure		Coolant		Lasers			
0.0 psi ⇌ +		OK		Off			
				1:	12.50 h		
				2:	0.00 h		

Label	Description
Control	Shows the voltage, current, and power.
Temperature	Shows the temperature of the cartridge and sample cooling system.
Tray	Shows the location of the capillary inlet and outlet.
LIF Detector	Shows information about the LIF detector. <ul style="list-style-type: none"> • Channels: The channel for the data, Ch1 and Ch2. • RFU: The relative fluorescence units of the data in this channel.
Pressure	Shows the direction and the magnitude of the pressure or vacuum. <ul style="list-style-type: none"> • ⇌: Forward direction • ⇐: Reverse direction • +: Pressure • -: Vacuum
Coolant	Shows the status of the coolant, OK or Low.
Lasers	(LIF detector) Shows the status of the laser, On or Off.

Direct Control

Label	Description
Laser Hrs	(LIF detector) Shows the number of hours that the laser has been on. <ul style="list-style-type: none">• 1: Hours for the integrated 488 nm laser.• 2: Hours for an external laser, if installed.

Figure 2-3 Instrument Status in the Direct Control Pane (PDA Detector)

Control	Temperature	Tray	PDA Detector
Voltage 0.000 kV	Cartridge 25.6 °C	BI: A1	Chan Wv Bw Absorb.
Current 0.000 µA	Storage 25.0 °C	BO: - -	Ch1 214 10 0.000000
Power 0 W		Lamp Hrs	Ch2 214 10 0.000000
Pressure 0.0 psi ➡ +	Coolant OK	D2: 7.00 h	Ch3 214 10 0.000000
	Lamp On	Hg: 0.06 h	Ref. 214 10 0.000000

Note: For items that are common to all detector types, refer to the figure: [Figure 2-2](#).

Label	Description
Lamp	Shows the status of the lamp, On or Off.
Lamp Hrs	Shows the number of hours that the lamps have been on. <ul style="list-style-type: none">• D2: The number of hours that the deuterium lamp has been on.• Hg: The number of hours that the mercury lamp has been on.
PDA Detector	Shows information about the PDA detector. <ul style="list-style-type: none">• Chan: The channel for the data.• Wv: The wavelength for the channel, in nm.• Bw: The bandwidth for the channel, in nm.• Absorb: The absorbance for the channel.

Figure 2-4 Instrument Status in the Direct Control Pane (UV Detector)

Control		Temperature		Tray		UV Detector	
Voltage	0.000 kV	Cartridge	25.2 °C	BI:	A1	Chan	Wv
Current	0.000 µA	Storage	25.0 °C	BO:	A1	Ch1	0
Power	0.000 W			Lamp Hrs		Absorb.	0.000000
Pressure	0.0 psi ➡ +	Coolant	OK	D2:	5.50 h		
		Lamp	On	Hg:	0.00 h		
							F

Note: For items common to all detector types, refer to the figure: [Figure 2-2](#).


Label	Description
Lamp	Shows the status of the lamp, On or Off.
Lamp Hrs	Shows the number of hours that the lamps have been on. <ul style="list-style-type: none"> D2: The number of hours that the deuterium lamp has been on. Hg: For display only. Not used for the UV detector.
UV Detector	Shows information about the UV detector. <ul style="list-style-type: none"> Chan: The channel for the data. Wv: The wavelength for the channel, in nm. Absorb: The absorbance for the channel.
F	Click to see filter information.

Figure 2-5 Instrument Status in the Direct Control Pane (UV Filters)

Control		Temperature		Tray		UV Filters	
Voltage	0.000 kV	Cartridge	25.2 °C	BI:	A1	F 1:	---
Current	0.000 µA	Storage	25.0 °C	BO:	A1	F 2:	200
Power	0.000 W			Lamp Hrs		F 3:	214
Pressure	0.0 psi ➡ +	Coolant	OK	D2:	5.50 h	F 4:	254
		Lamp	Off	Hg:	0.00 h	F 5:	280
							D

Note: For items common to all detector types, refer to the figure: [Figure 2-2](#).

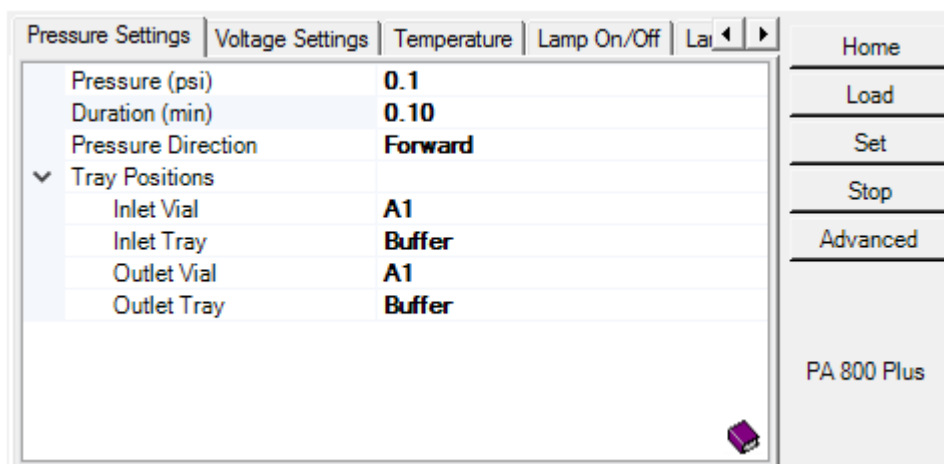
Direct Control

Label	Description
Lamp	Refer to the figure: Figure 2-4 .
Lamp Hrs	Refer to the figure: Figure 2-4 .
UV Filters	F<x> : Shows the wavelength of the filter in position <x>, in nm.
	Click to see detector information.




Parameters and Buttons in the Direct Control Pane

Note: Pressure values can be shown in millibar (mbar) or pounds per square inch (psi). The registry setting for the Waters Empower™ software controls which value is used. The default unit is millibar. To change the units, refer to the document: *PA 800 Plus Empower™ Driver Release Notes*.

Figure 2-6 Parameters and Buttons in the Direct Control Pane



Label	Description
Parameter Tabs	
Pressure Settings	Set the pressure for the system.
Voltage Settings	Set the voltage for the system.
Temperature	Set the temperature for the capillary and sample cooler.

Label	Description
Lamp On/Off	(UV or PDA detector) Turn the lamp on or off.
Laser On/Off	(LIF detector) Turn the laser on or off.
Calibration Factors	(LIF detector) See the calibration correction factors and set the parameters for detector calibration. Refer to the section: Calibrate the LIF Detector (Optional) .
UV Filters	(UV detector) Set the position and wavelength of the filters installed in the system.
Lamp Hours	(UV or PDA detector) After the lamp is replaced, set the lamp hours to 0.
Lamp Energy	(UV detector) Select the filter in the Filter list, and then click Set to see the current between the diodes in the deuterium lamp, in nA. This value decreases over time as the lamp ages.
Trays	See the type of sample and buffer trays in use.
Spectrum	(PDA detector) See the spectrum of the deuterium lamp. Refer to the section: See the Deuterium Lamp Spectrum and Intensity .
Buttons	
	Click to open the next or previous tab.
	Click to open the Help pane.
	Click to close the Help pane.
Home	Click to move the trays to the home position.
Load	Click to move the trays to the load position.
Set	<p>Click to send the parameters to the PA 800 Plus system.</p> <ul style="list-style-type: none"> • (LIF detector) When the Calibration Factors tab is shown, this button changes to Start. • (UV detector) When the Lamp Hours tab is shown, this button changes to Reset. • (PDA detector) When the Spectrum tab is shown, this button changes to Monitor.

Direct Control

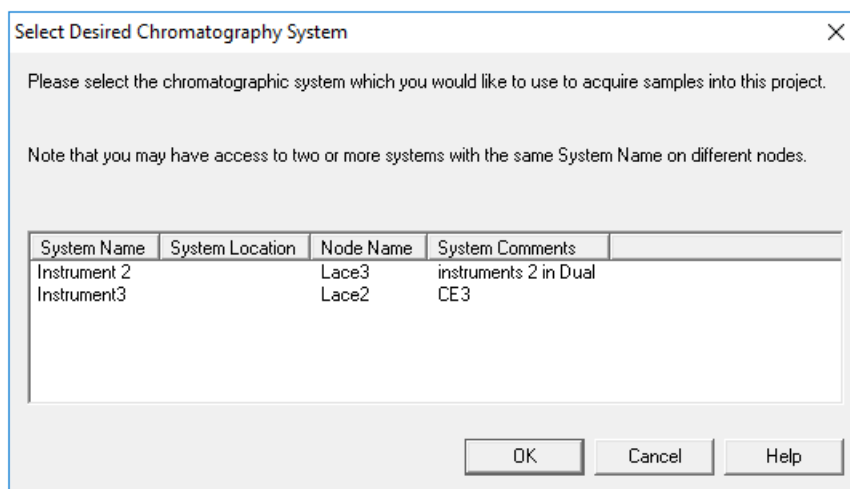
Label	Description
Stop	Click to turn off the voltage, current, power, pressure, and cooling.
Advanced	(PDA detector) Click to calibrate the PDA detector. Refer to the section: Calibrate the PDA Detector .

Create an Instrument Method

3

1. In the Waters Empower™ software Project window, click **File > New Method > Instrument Method**.

Figure 3-1 Select Desired Chromatography System Dialog



2. Click the applicable system name, and then click **OK**.
3. In the Instrument Method Editor, do this:
 - a. Open the Detector tab.
 - b. From the **Detector Type** list, select the applicable detector.
 - c. Set the parameters.
Refer to the section: [Detector Parameters for an Instrument Method](#).
 - d. Make sure that the system is configured with the correct detector for the application. If the **Detector Type** must be changed, then change it before other changes are made to the instrument method.

Note: When the **Detector Type** changes, all parameters are set to their default values.

Create an Instrument Method

Figure 3-2 Detector Parameters

General | **Detector** | Time Program

Detector Type: PDA

Electropherogram Scan Data

Data Rate: 4 Hz

Scan Range from: 190 to 300 nm

Filter: General Purpose 16-25

Electropherogram Channel Data

Data Rate: 4 Hz

	Acquire	Ref	Wl [nm]	Bw [nm]
Channel 1	<input type="checkbox"/>	<input type="checkbox"/>	214	10
Channel 2	<input type="checkbox"/>	<input type="checkbox"/>	254	10
Channel 3	<input type="checkbox"/>	<input type="checkbox"/>	280	10
Peak Detect.	<input type="checkbox"/>	<input type="checkbox"/>	250	120

Relays

Relay 1: Closed

Relay 2: Closed

Reference Channel

Wavelength: 400 nm

Bandwidth: 10 nm

Absorbance Signal

Signal: Direct

4. Open the General tab, and then set the parameters. Refer to the section: [General Parameters for an Instrument Method](#).

Figure 3-3 General Parameters

General | Detector | Time Program

Auxiliary Data Channels

☐ Voltage Max: 30.0 kV

☒ Current Max: 300.0 μ A

Power Max: 9.000 W

☐ Pressure

☐ Cartridge Temperature

Peak Detect Parameters

Peak Noise Multiplier 2

Peak Filter Width 9

Capillary Settings

Capillary Total Length 60.2 cm

Capillary Length 50.0 cm

Trigger Settings

☐ Wait For External Trigger

Wait for Temperature

Do not wait

Temperature

Cartridge 25.0 $^{\circ}$ C

Sample Storage 25.0 $^{\circ}$ C

Inlet Trays

Buffer 36 vials

Sample 48 vials

Outlet Trays

Buffer 36 vials

Sample No tray

5. To add events to the time program, open the Time Program tab. Refer to the section: [Add Events to the Time Program for an Instrument Method](#).

The Waters Empower[™] software requires that the last event in the time program is an **End** event.

Figure 3-4 Time Program

General		Detector		Time Program					
	Time (min)	Event	Value	Duration	Inlet vial	Inlet tray	Outlet vial	Outlet tray	Summary
▶		Rinse Pressure ▾	20.0 psi	2.00 min	A1	Buffer	A1	Buffer	Forward;0;0
	0.00	Separate Pre... ▾	20.0 psi	2.00 min	B1	Buffer	B1	Buffer	Forward;0;0
	0.20	Autozero ▾							
	2.00	End ▾							
*		▾							

6. To save the instrument method, do this:
- Click **File > Save**.
The Save current Instrument Method dialog opens.
 - In the **Name** field, type a name.
 - (Optional) In the **Method Comments** field, type any required information.

Create an Instrument Method

- d. If a prompt is shown, then in the **Password** field, type the Waters Empower™ software password for the current user, and then click **Save**.

The instrument method is saved to the active project.

General Parameters for an Instrument Method

Figure 3-5 General Parameters for an Instrument Method

General | Detector | Time Program

Auxiliary Data Channels

☐ Voltage Max: 30.0 kV

☒ Current Max: 300.0 μ A

☐ Power Max: 9,000 W

☐ Pressure

☐ Cartridge Temperature

Peak Detect Parameters

Peak Noise Multiplier 2

Peak Filter Width 9

Capillary Settings

Capillary Total Length 60.2 cm

Capillary Length 50.0 cm

Trigger Settings

☐ Wait For External Trigger

Wait for Temperature

Do not wait

Inlet Trays

Buffer 36 vials

Sample 48 vials

Outlet Trays

Buffer 36 vials

Sample No tray

Temperature

Cartridge 25.0 $^{\circ}$ C

Sample Storage 25.0 $^{\circ}$ C

Label	Description
Auxilliary Data Channels	Select more types of data to be collected: Voltage , Current , Pressure , and Cartridge Temperature . For Voltage , Current , and Power , type the maximum value to be applied during data collection.

Label	Description
Trigger Settings	<p>If the method is triggered by an outside source or device, then select the Wait For External Trigger check box.</p> <p>To use a temperature to start the run, select an option: Do not wait, Wait for Cartridge Temperature, Wait for Storage Temperature, and Wait for Cartridge and Storage Temperature.</p>
Inlet Trays	Select the type of sample and buffer trays that are installed in the inlet positions.
Peak Detect Parameters	Do not change the parameters in this area. They have no effect on data acquisition.
Capillary Settings	Type the dimensions of the capillary.
Temperature (°C)	Type the temperature for the cartridge and the sample cooler.
Outlet Trays	Select the type of sample and buffer trays that are installed in the outlet positions.

Detector Parameters for an Instrument Method

Figure 3-6 Detector Parameters for a PDA Detector

General

Detector

Time Program

Detector Type

PDA

Electropherogram Scan Data

Data Rate

4

Hz

Scan Range from

190

to

300

nm

Filter

General Purpose 16-25

Electropherogram Channel Data

Data Rate

4

Hz

	Acquire	Ref	Wl [nm]	Bw [nm]
Channel 1	<input type="checkbox"/>	<input type="checkbox"/>	214	10
Channel 2	<input type="checkbox"/>	<input type="checkbox"/>	254	10
Channel 3	<input type="checkbox"/>	<input type="checkbox"/>	280	10
Peak Detect.	<input type="checkbox"/>	<input type="checkbox"/>	250	120

Relays

Relay 1

Closed

Relay 2

Closed

Reference Channel

Wavelength

400

nm

Bandwidth

10

nm

Absorbance Signal

Signal

Direct

Label	Description
Detector Type	Select the type of detector.
Electropherogram Scan Data	<div>Set the sampling rate of the data to be collected, in Hz, and the wavelength range to be scanned, in nm.</div> <div>A higher rate increases the number of data points per peak, but might cause more noise. The optimal rate is different for each analyte and is usually found during method development.</div> <div>Note: The value for Data Rate must be between 25% to 100% of the Data Rate for the Electropherogram Channel Data.</div>

Label	Description
Electropherogram Channel Data	<p>Set the parameters for data collection for as many as three channels. In the Data Rate field, select the sampling rate for data collection.</p> <p>A higher rate increases the number of data points per peak, but might cause more noise. The optimal rate is different for each analyte and is usually found during method development.</p> <p>For each channel, do this:</p> <ul style="list-style-type: none">• To acquire data from this channel, select the Acquire check box.• To subtract reference trace data from the collected data, select the Ref check box. The reference is a wavelength that is recorded and subtracted from the data in the wavelength channel.• Type the Wavelength of the data to be collected, in nm.• Type the Bandwidth of the data to be collected, in nm.
Filter	<p>Select the filter to use to decrease noise in the data.</p> <p>Refer to the section: About the Filter Parameter.</p>
Relays	For Relay 1 and Relay 2 , select Open or Closed .
Reference Channel	Type the values for the Wavelength and Bandwidth for the reference channel, in nm.
Absorbance Signal	<p>To show the data as received from the detector, select Direct.</p> <p>To invert the signal before the data is shown, select Indirect.</p>

Create an Instrument Method

Figure 3-7 Detector Parameters for a LIF Detector

The screenshot shows the 'Detector' tab of a software interface. At the top, there are three tabs: 'General', 'Detector', and 'Time Program'. The 'Detector' tab is selected. Below the tabs, there is a 'Detector Type' dropdown menu set to 'LIF'. The main area is divided into two columns for 'Electropherogram Channel 1' and 'Electropherogram Channel 2'. Channel 1 settings include: 'Acquisition enabled' (checked), 'Dynamic range' (100 RFU), 'Filter' (General Purpose 16-25), 'Fluorescence Signal' (Direct), 'Excitation wavelength' (488 nm), 'Emission wavelength' (520 nm), and 'Data rate' (2 Hz). Channel 2 settings include: 'Acquisition enabled' (unchecked), 'Dynamic range' (100 RFU), 'Filter' (General Purpose 16-25), 'Fluorescence Signal' (Direct), 'Excitation wavelength' (635 nm), 'Emission wavelength' (675 nm), and 'Relays' (Relay 1: Closed, Relay 2: Closed).

Label	Description
Detector Type	Select the type of detector.
Acquisition enabled	Select to enable data acquisition for the channel. Data can be acquired from one or both channels.
Acquisition	Select the upper limit of the data to be collected, in RFU. If the fluorescence signal is above this limit, then peaks might be truncated.
Filter	Select the filter to use to decrease noise in the data. Refer to the section: About the Filter Parameter .
Fluorescence Signal	To show the data as received from the detector, select Direct . To invert the signal before the data is shown, select Indirect .

Label	Description
Laser/filter description - information only	Type the values for the excitation and emission wavelengths, in nm. These values are kept with the method but are not used for acquisition. The laser wavelength and the emission filter installed in the LIF detector control the excitation and emission wavelengths that are used for data acquisition.
Data rate	For both channels, select the sampling rate for the LIF data to be collected, in Hz. A higher rate increases the number of data points per peak, but might cause more noise. The optimal rate is different for each analyte and is usually found during method development.
Relays	For Relay 1 and Relay 2 , select Open or Closed .

Figure 3-8 Detector Parameters for a UV Detector

General | **Detector** | Time Program

Detector Type: UV

Filter: General Purpose 16-25

Electropherogram Channel Data

Data Rate: 4 Hz

Wavelength: 210 nm

Relays

Relay 1: Closed

Relay 2: Closed

Absorbance Signal

Signal: Direct

Create an Instrument Method

Label	Description
Detector Type	Select the type of detector.
Electropherogram Channel Data	Type the values for the Data Rate , in Hz, and the Wavelength , in nm, for data collection. A higher rate increases the number of data points per peak, but might cause more noise. The optimal rate is different for each analyte and is usually found during method development.
Filter	Select the filter to use to decrease noise in the data. Refer to the section: About the Filter Parameter .
Relays	For Relay 1 and Relay 2 , select Open or Closed .
Absorbance Signal	To show the data as received from the detector, select Direct . To invert the signal before the data is shown, select Indirect .

About the Filter Parameter

The following types of noise filters are available. For each type of filter, a peak width can be specified.

- **General Purpose:** This is the normal noise filter. It maximizes peak smoothing with limited or minimal peak distortion and loss of resolution.
- **Max Sensitivity:** This filter decreases baseline noise. It maximizes the signal-to-noise ratio, but might cause peaks to broaden or flatten. Use this for experiments where peaks are resolved and detection limits or quantitative accuracy is most important.
- **Max Resolution:** This filter keeps the peak shape, but decreases baseline noise less than the other filter options.

The peak width is the expected peak width at the base of a peak. The following ranges are available:

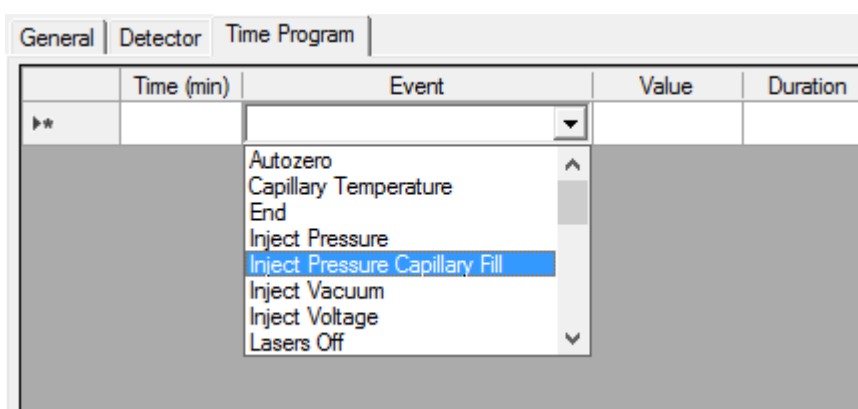
- **None:** No filtering is done.
- **<16 points:** The noise filter uses the smallest number of points. The result is less smoothing and more noise.
- **16 - 25 points:** The noise filter uses an intermediate number of points.
- **>25 points:** The noise filter uses the largest number of points. The result is more smoothing and less noise.

Add Events to the Time Program for an Instrument Method

The time program is a table of events in an instrument method. The events are done in order, from top to bottom.

1. Open an instrument method, and then open the Time Program tab.
2. Click the **Event** list, and then select an event. Refer to the table: [Table A-1](#).

Figure 3-9 Event List in the Time Program Tab



Fields for the event parameters are shown in the pane below the table.



3. As required, in the fields to the right, type the applicable parameter values. Refer to the table: [Table A-2](#).

Figure 3-10 Edit Event Parameters on the Time Program Tab

Pressure (psi)	25.0
Duration (s)	100.0
Pressure Direction	Forward
▼ Tray Positions	
Inlet Vial	A1
Inlet Tray	Buffer
Outlet Vial	A1
Outlet Tray	Buffer
▼ Increment Every Runs[]	
Inlet	0
Outlet	0
Comments	

Create an Instrument Method

Tip! If a value cannot be changed, then it might be in a list. To make a selection from the list, scroll to the far right to show the **Down** arrow, and then click the arrow.

4. (Optional) To show the applicable ranges for the parameters, click .
To hide the Help pane, click .
5. (Optional) To add more rows to the time program, right-click a row header, and then select **Insert Row**.
The new row is added below the selected row.
6. (Optional) To delete rows from the time program, right-click a row header, and then select **Remove Row**.
The row is deleted.
7. If this time program includes any of the Separate events, then add the **End** event as the last event in the time program.
8. Save the instrument method:
 - a. To open the Save current Instrument Method dialog, click **File > Save**.
 - b. In the **Name** field, type a name.
 - c. (Optional) In the **Method Comments** field, type any required information.
 - d. If a prompt is shown, then in the **Password** field, type the Waters Empower™ software password for the current user, and then click **Save**.

The instrument method is saved to the current project.

Set the Definition for the Buffer and Sample Trays

4

In the Waters Empower™ software, the sample and buffer trays in the PA 800 Plus system are referred to as *plates*. The plate definition must be set in the Waters Empower™ software. To make this process easier, SCIEX supplies the required information in text files that can be imported.

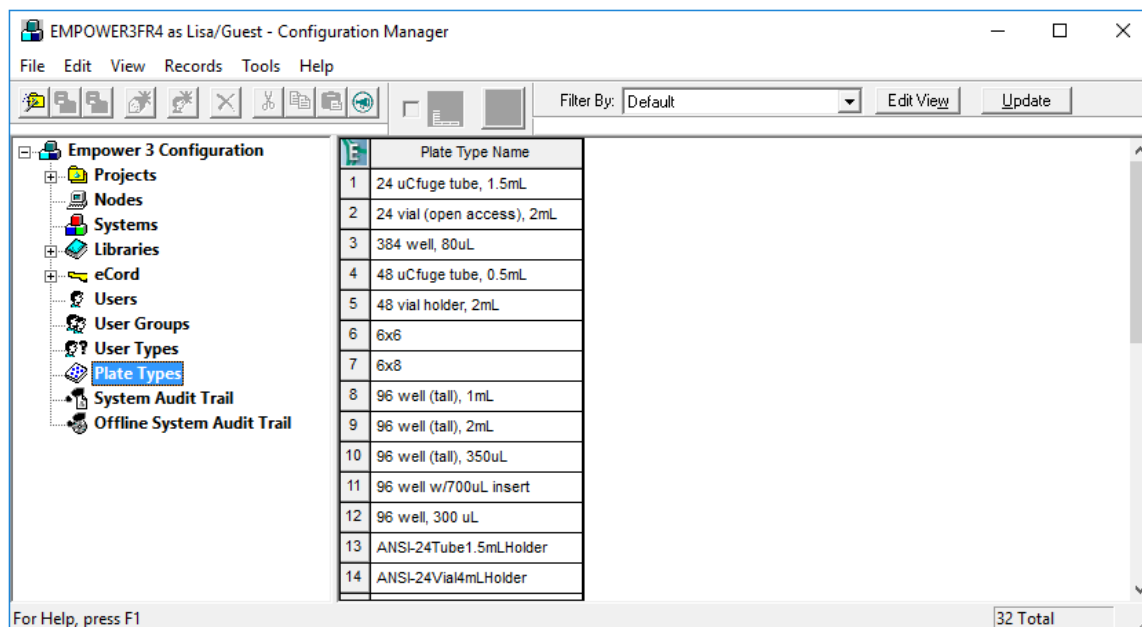
Note: The plate definitions should have been set when the Waters Empower™ software was installed. If the list of plates in the Plate Types Name table includes the PA 800 Plus Sample Tray, PA 800 Plus Buffer Tray, and PA 800 Plus 96 Well Sample Tray, then the plate definitions have already been set. The procedure is included here for reference.

1. Put the PA 800 Plus Empower™ Driver DVD in the DVD drive.

Note: If the DVD is not available, then use the copies of the files that are included in this document. Refer to the section: [Plate Definition Files](#). For each file, copy the contents, and then paste the contents in an individual text file.

2. Click **Configure the System**.

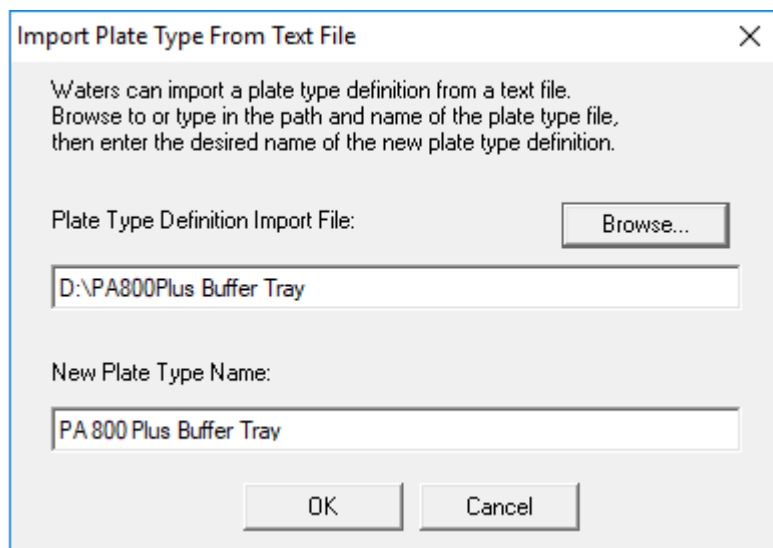
Figure 4-1 Configuration Management Window: Plate Types



Set the Definition for the Buffer and Sample Trays

3. To show the plates definitions that are already set, click **Plate Types**.
4. Create the plate definition for the buffer tray:
 - a. Right-click the table, and then select **Import from Text**.

Figure 4-2 Import Plate Type From Text File Dialog



- b. Click **Browse**, and then find and click the `PA800Plus Buffer Tray.txt` file.
 - c. In the **New Plate Type Name** field, type `PA 800 Plus Buffer Tray`, and then click **OK**.

The buffer tray is added to the list in the Configuration Manager window.

5. To create the plate definition for the sample trays, do step 4 again.
 - For the 48-vial sample tray, find and click the `PA800Plus Sample Tray.txt` file. In the **New Plate Type Name** field, type `PA 800 Plus Sample Tray`.
 - For the 96-well sample tray, find and click the `PA800Plus 96 Well Sample Tray.txt` file. In the **New Plate Type Name** field, type `PA 800 Plus 96 Well Sample Tray`.

Note: The plate definition file for the 96-well sample plate is for a standard SCIEX 96-well plate (PN 609844). To use a 96-well plate from another manufacturer, in the Configuration Manager window, click **File > New > Plate Type**, and then set the plate definition manually.

6. If the Beckman Coulter PACE MDQ Control for Waters Empower™ Software Driver was previously installed, then delete any plates that were created for use with the driver. Right-click the row number for the plate, and then select **Delete**.

Set the Definition for the Buffer and Sample Trays

7. (Optional) To see detailed information about a plate, right-click the row number for the plate, and then select **Properties**.
8. (Optional) To delete a plate, right-click the row number for the plate, and then select **Delete**. Only plates that were added by a user can be deleted. Plates with preset definitions cannot be deleted.
9. To close the **Configuration Manager**, click **File > Exit**.

This section gives instructions to use the Waters Empower™ software to change the UV lamp and to calibrate the PDA and LIF detectors.

For more maintenance procedures, refer to the document: *Maintenance Guide*.

Change the Detector

1. In the Waters Empower™ software, close the Run Samples window.
2. To open the Configuration Manager window, in the Waters Empower™ software Start dialog, click **Configure the System**.
3. To show the available nodes, click **Node** in the Empower Configuration tree.
4. Click the row number for the applicable node, and then right-click **Bring Offline**.
If the system is not in use, that is, if no users are connected to the system or no samples are being acquired, then the system goes offline. If the system is in use, then a message is shown.
5. Close any open programs, and then start the LAC/E module again.
6. Change the detector. Refer to the document: *Maintenance Guide*.
For a UV detector, make a note of the positions of any filters that are installed in the UV source optics assembly.
7. In the Configuration Manager window, click the row number for the applicable node, and then right-click **Bring Online**.
8. To close the message dialog, click **OK**.
9. Do one of the following:
 - For a PDA or LIF detector, calibrate the detector. Refer to the section: [Calibrate the PDA Detector](#) or [Calibrate the LIF Detector \(Optional\)](#).
 - For a UV detector, continue with step 10.
10. (UV detectors only) Set the filter information:
 - a. In the Direct Control window, click **F**, and then open the **UV Filters** tab.
 - b. For each position in the detector where there is a filter, type the wavelength of the filter.
The following table shows the default values.

Table 5-1 Default Filter Wavelengths for the UV Detector

Position	Wavelength
Filter Position 2	200
Filter Position 3	214
Filter Position 4	254
Filter Position 5	280

- c. Click **Set**.

See the Deuterium Lamp Spectrum and Intensity

Use this procedure to see the raw counts from the deuterium lamp as seen by the detector. If the signal is low, then this procedure can find if the low signal is caused by an issue with the lamp.

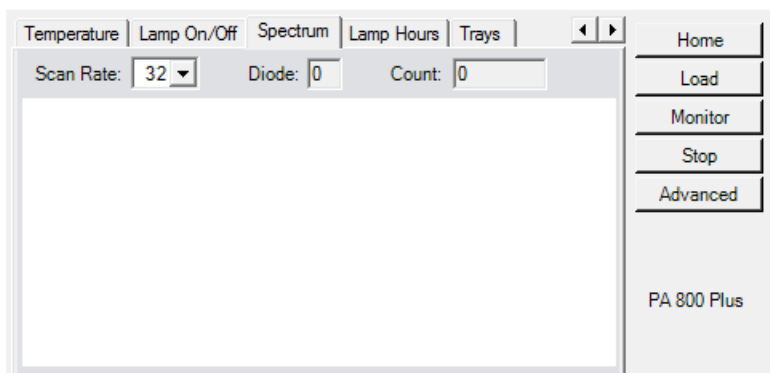
The spectrum is a better indicator of lamp life than the **Lamp Hours** value.

Required Materials

- PDA detector
- OPCAL cartridge (PN 144660)

1. Install the PDA detector. Refer to the section: [Change the Detector](#), and the document: *Maintenance Guide*.
2. In the **Direct Control** pane, open the **Lamp On/Off** tab.
3. To turn on the lamp, click **On**, and then click **Set**.
4. Open the Spectrum tab. In the **Scan Rate** list, select **32**, and then click **Monitor**.

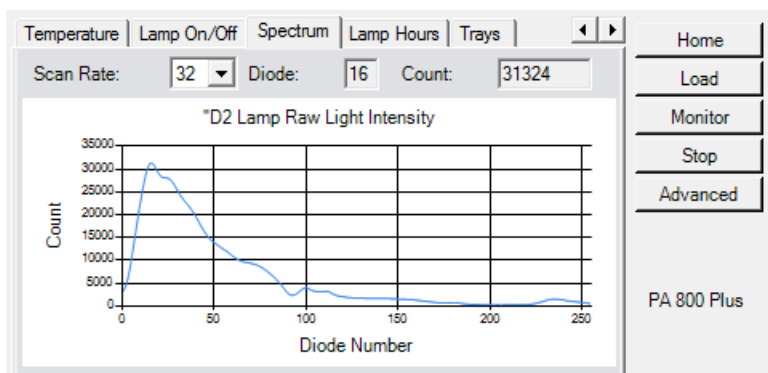
Figure 5-1 Spectrum Tab



System Maintenance

When the data is collected, the spectrum is shown.

Figure 5-2 Spectrum Tab with Acceptable Spectrum

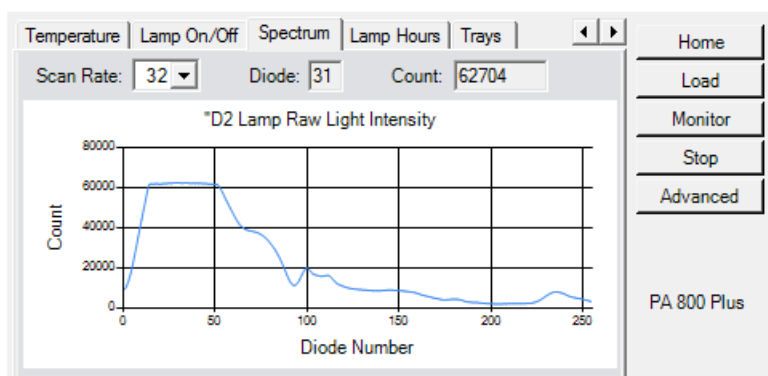


5. Examine the spectrum and the value in the **Counts** field.

- If the value is more than 5,000 and the plot is not flat on top, then the lamp operation is correct.
- If the value is less than 5,000, then go to step 6.
- If the plot is flat on top, then the signal is saturated. In the **Scan Rate** list, select **64**, and then click **Monitor**.

If the plot is still flat, then in the **Scan Rate** list, select **128**, and then click **Monitor**.

Figure 5-3 Spectrum Tab with Saturated Spectrum



6. Examine the cartridge to make sure that:

- The aperture is clean.
- The capillary is clean and not broken.
- The aperture is in the center of the capillary window.

- The fiber-optic cable is clean and not broken.
7. In the **Scan Rate** list, select **32**, and then click **Monitor**.
In the **Counts** field, if the value is still less than 5,000 at 32 Hz, then go to step 8.
 8. Install the OPCAL cartridge.
 9. In the **Scan Rate** list, select **32**, and then click **Monitor**.
In the **Counts** field, if the value is less than 10,000, then replace the lamp. Refer to the section: [Change the Deuterium Lamp](#).

Change the Deuterium Lamp

Required Materials

- Deuterium lamp
- 7/64 inch hex key
- Powder-free gloves



WARNING! Hot Surface Hazard. Before a lamp is replaced, turn off the power to the system, and then let the lamp cool fully. A hot lamp will cause burns.

CAUTION: Potential Wrong Result. Before the deuterium lamp is installed, make sure that an orange O-ring is installed on the lamp flange. A missing O-ring decreases lamp performance.

CAUTION: Potential System Damage. Do not touch the envelope of the lamp. Oils from skin can cause damage to the lamp.

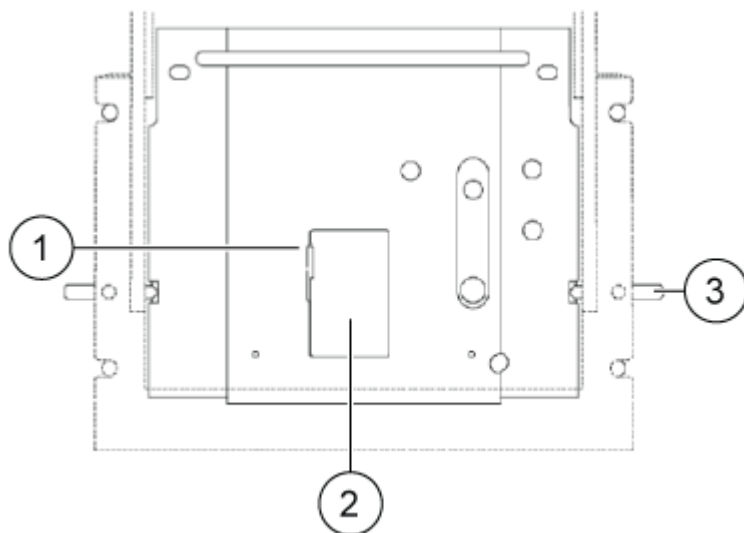
CAUTION: Potential System Damage. Use powder-free gloves to touch the UV lamp. UV lamp operation generates high temperatures and strong UV intensity. In these conditions, fingerprints make corrosive compounds that etch the surface of the UV lamp and can cause it to break when it is turned on. Keep the UV optical window dry and protect it from abrasion.

The deuterium lamp is used by the UV and PDA detectors. If the baseline shows too much noise or the lamp does not illuminate, then the deuterium lamp might need to be replaced.

1. Open the cartridge cover.
2. Turn off the power to the system, and then let the deuterium lamp cool.
3. Loosen the two thumbscrews on the insertion bar, and then lift the insertion bar.
4. Remove the capillary cartridge from the interface block.

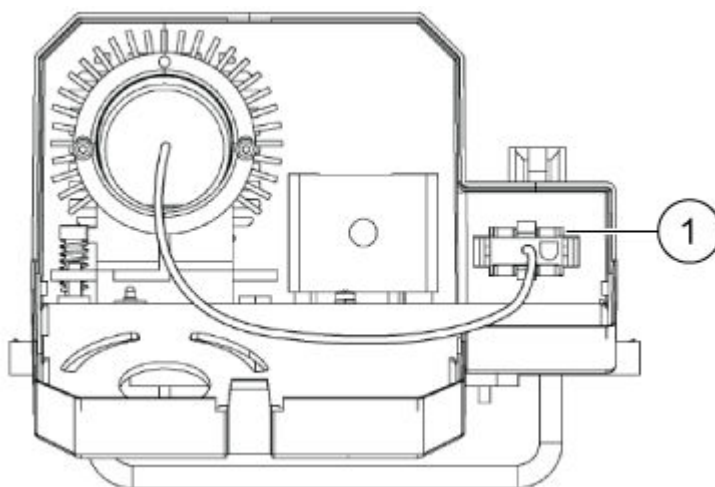
5. Loosen the two thumbscrews on the optics source assembly.

Figure 5-4 Optics Source Assembly



Item	Description
1	Access cover latch
2	Access cover
3	Thumbscrews (one on each side)

6. Pull forward to remove the optics source assembly.
7. Put the assembly on a clean work surface.
8. Open the access cover on the back of the optics source assembly, and then disconnect the power connector.

Figure 5-5 Deuterium Lamp Assembly

Item	Description
1	Power connector

9. Use a 7/64 inch hex key to remove the two screws that hold the deuterium lamp in position, and then remove the lamp from the lamp housing.
10. To install the new deuterium lamp, align the flange guide notch in the lamp with the housing guide pin.
11. Install the two hex screws, and then tighten them.
12. Connect the power plug to the optics source assembly, and then close the access cover.
13. Put the optics source assembly in the mounting location, align the two upper guide pins, and then tighten the two thumbscrews.
14. Install the capillary cartridge in the interface block.
15. Lower the insertion bar, and then tighten the two thumbscrews.
16. Close the cartridge cover.
17. Turn on the power to the system.
18. Reset the lamp hours in the Waters Empower™ software:
 - a. Start the Waters Empower™ software.
 - b. In the Direct Control pane, click **Lamp Hours**, and then click **Reset**.

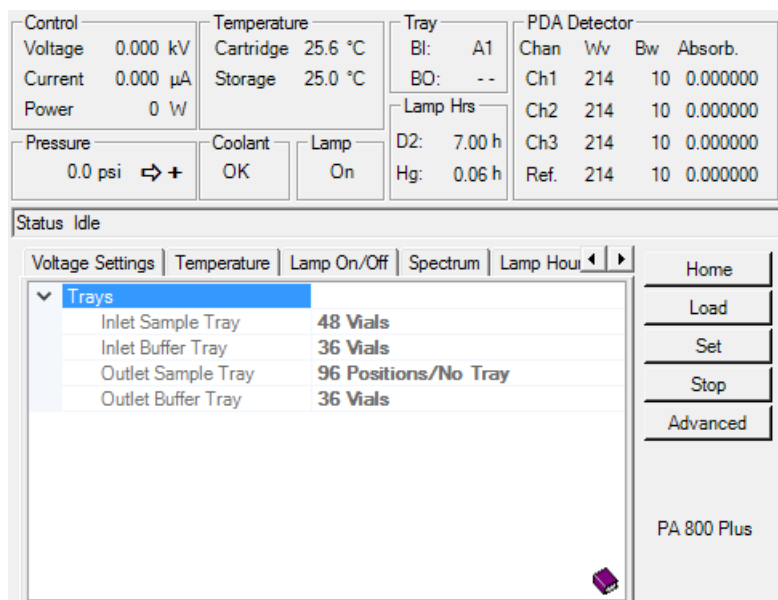
Calibrate the PDA Detector

To make sure that the analysis results are consistent over time, calibrate the PDA detector after the PDA detector is installed, after a different cartridge is installed, and after a new capillary is installed in the cartridge.

1. Turn off the PA 800 Plus system.
2. Install the PDA detector.
Refer to the document: *Maintenance Guide*.
3. Turn on the PA 800 Plus system, and then let the lamp warm up for at least 30 minutes.
4. Open the Waters Empower™ software, and then click **Run Samples**.
The Direct Control pane shows in the Run Samples window.

Note: If the Direct Control pane does not show, then click **View > Control Panels > SCIEX CE**.

Figure 5-6 Direct Control Pane for PDA Detector



5. In the Direct Control pane, click **Advanced**.
The window changes to show more parameters.

Figure 5-7 PDA Detector Calibration Parameters

Electropherogram Channel Data			
	Ref	Wl [nm]	Bw [nm]
Channel 1	<input type="checkbox"/>	214	10
Channel 2	<input type="checkbox"/>	254	10
Channel 3	<input type="checkbox"/>	280	10
Peak Detect.	<input type="checkbox"/>	250	120

Absorbance Signal
 Direct

Filter
 General Purpose 16-25

Shutter
 Closed

Reference Channel
 Wavelength: 400 nm
 Bandwidth: 10 nm

Apply
 Cancel
 Calibrate

Status:

6. Click **Calibrate**. Do not change any of the parameters.
The calibration starts. When the calibration is complete, the status field shows 87: PDA Wavelength calibration successful!, where 87 is the message code.
7. If the calibration is unsatisfactory, then do this:
 - a. Remove the cartridge and detector.
 - b. Install the cartridge and detector again.
 - c. Do step 3 through step 6.

If the calibration is unsatisfactory a second time, then do this step again.

If the calibration is unsatisfactory again, then contact SCIEX Technical Support at sciex.com/request-support.

Calibrate the LIF Detector (Optional)

This procedure is optional. If there is a requirement for consistency from system to system or cartridge to cartridge, then do this procedure.

Calibrate the LIF detector after the LIF detector is installed, after a different cartridge is installed, or after a new capillary is installed in the cartridge.

Note: The following procedure technically does normalization, not calibration. Normalization uses a measured quality, such as the fluorescence of the LIF Performance Test Mix. Calibration uses an external standard. Because the software user interface uses the term *calibration*, that term is used in this guide.

For the best results, we recommend the use of the following:

System Maintenance

- The same capillary that will be used for the separations
- For each system, a dedicated LIF aperture and probe guide

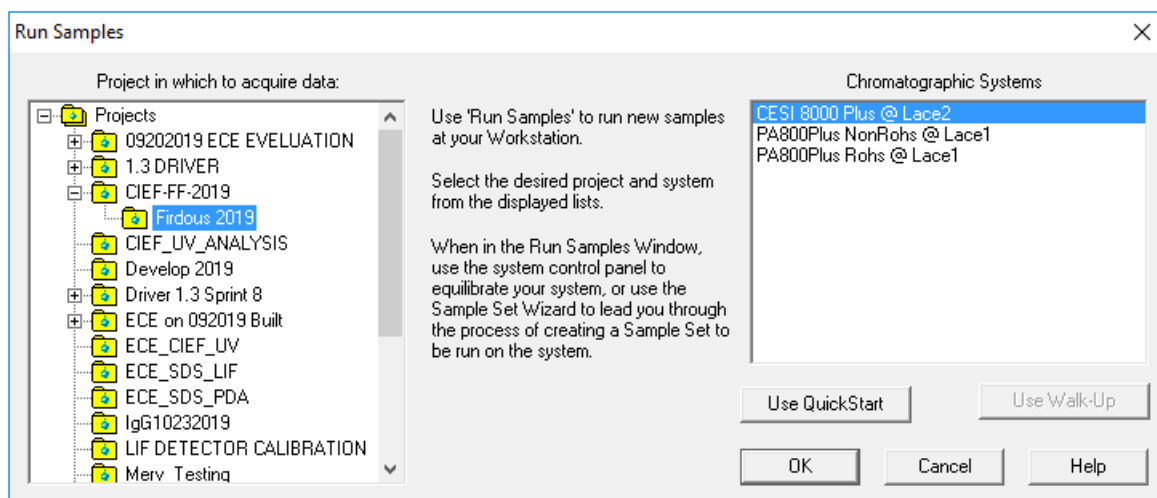
Required Materials
<ul style="list-style-type: none">• LIF Performance Test Mix• CE Grade Water

1. Turn off the PA 800 Plus system.
2. Install the LIF detector.
3. Turn on the PA 800 Plus system, and then turn on the solid-state laser.
4. Open the Waters Empower™ software.

Figure 5-8 Waters Empower™ software Pro Interface Window



5. Click **Run Samples**. If required, then log in.

Figure 5-9 Run Samples Dialog

6. On the left, click the folder for the project of interest.
7. On the right, click the system that has the LIF detector installed, and then click **OK**.
8. In the Direct Control pane, click **Load**.
9. In the buffer outlet tray, put a universal vial in position A1.
10. In the buffer inlet tray, put universal vials in positions A1 and B1.

Note: To prevent splashing, put the empty vials in the tray, and then add liquid and attach the caps.

11. Fill the vials, and then put caps on the vials:
 - Inlet buffer tray position A1: 1.5 mL of CE Grade Water
 - Inlet buffer tray position B1: 1.5 mL of LIF Performance Test Mix
 - Outlet buffer tray position A1: 1.0 mL of CE Grade Water
12. Set the parameters, and then start the calibration:
 - a. In the Direct Control pane, open the Calibration Factors tab, and then click **Auto**.

Figure 5-10 Direct Control Pane: Calibration Factors Tab

Voltage Settings | Temperature | Lasers On/Off | Calibration Factors

Please enter the following calibration parameters:

Detector channel: ☒ 1 ☐ 2

Target RFU value: RFU

Capillary dimensions

Internal diameter: µm

Total length: cm

☐ Manual ☒ Auto

PA 800 Plus

Home Load Start Stop

- b. Click the **Detector channel** to be calibrated.
- c. In the **Target RFU value** field, type the recommended RFU value. Refer to the following table.

Table 5-2 Recommended Target RFU by Capillary Type

Type of Capillary	Internal Diameter (µm)	Total Length (cm)	Recommended Target RFU
Bare fused-silica	50	User specified	15
Bare fused-silica	75	User specified	35
DNA	100	User specified	62
N-CHO	50	User specified	7

- d. Type the values for the **Internal diameter** and **Total length** of the capillary.
- e. Click **Start**.

Figure 5-11 Vial positions Dialog

Vial positions

You are about to start LIF Calibration. Please check the appropriate vials are in the following positions:

Inlet A1: Buffer

Inlet B1: Calibration Mix

Outlet A1: Waste

Do you want to continue?

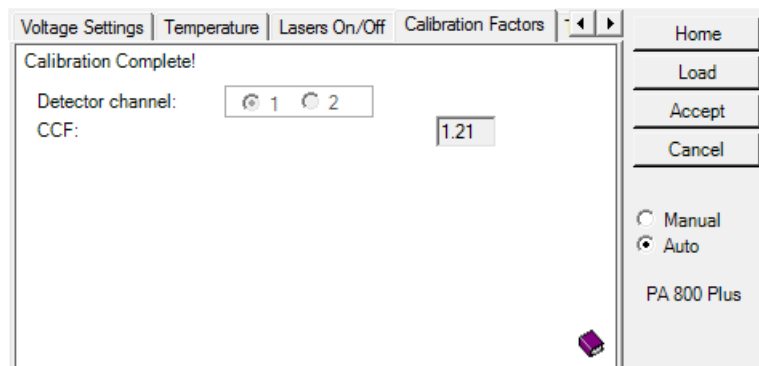
Yes No

f. Click **Yes**.

The calibration starts and requires approximately 9 minutes to complete. The Calibration Complete! message is shown.

If the message No step change detected is shown, then the detector cannot detect the solution. For troubleshooting procedures, refer to the section: [No Step Change Detected](#).

Figure 5-12 Calibration Factors Tab After Calibration



13. Examine the value in the **CCF** field:

- If the CCF value is less than 0.1, then click **Cancel**. Refer to the section: [CCF Values for LIF Detector Calibration](#).
- If the CCF value is between 0.1 and 10, then the calibration was successful. Click **Accept** to save the results.

Note: If the samples will be labeled with a dye other than fluorescein, then we recommend that a standard be run to make sure that the system performance is satisfactory.

- If the CCF value is more than 10, then click **Cancel**. Refer to the section: [CCF Values for LIF Detector Calibration](#).

14. If the system will be used immediately for separation, then on the Temperature tab, set the sample storage temperature to a value that is correct for the samples to be run.

By default, after the calibration completes, the sample storage temperature is set to 25 °C.

15. Close the Direct Control pane.

Troubleshoot the LIF Detector Calibration

CCF Values for LIF Detector Calibration

Issue	Action
Reported CCF value is less than 0.1 or System performance is not satisfactory	<ul style="list-style-type: none"> • Make sure that the correct capillary was used, and that it is not broken. • Make sure that the laser output for the laser in use on the system is correct. • Make sure that the correct filters are installed in the LIF detector: <ul style="list-style-type: none"> • Excitation: 488 nm • Emission: 520 nm • Replace the test mix, buffer, and capillary, and then do the calibration again. If the issue continues, then contact SCIEX Technical Support at sciex.com/request-support.
Reported CCF value is between 0.1 and 10.0	There is no issue with the system. Run a standard and make sure that the system performance is satisfactory.
Reported CCF value is more than 10 or System performance is not satisfactory	<ul style="list-style-type: none"> • Make sure that the laser output for the laser in use on the system is correct. • Make sure that the correct filters are installed in the LIF detector: <ul style="list-style-type: none"> • Excitation: 488 nm • Emission: 520 nm • Replace the test mix, buffer, and capillary, and then do the calibration again. If the issue continues, then contact SCIEX Technical Support at sciex.com/request-support.

No Step Change Detected

The LIF calibration compares detector signals from a nonfluorescent solution and a known fluorescent solution. When a rinse with nonfluorescent solution is done and then followed by a rinse with fluorescent solution, the first part of the detector signal should be near zero and the second part should be near the target fluorescent value. This detector output is in the shape of a step and is referred to as a *step change*. If a step change is not seen, then the applicable solutions are not passing the detector or the detector cannot detect the solutions.

1. Make sure that the switch on the right side of the laser is in the ON position.
2. Make sure that the laser that was supplied with the system is connected and the LASER ON light is illuminated.

3. To make sure that the solution goes through the capillary, from buffer inlet position A1 to an empty buffer vial in outlet position B1, use Direct Control to do a pressure rinse with CE Grade Water at 20 psi for 5 minutes.
4. When the rinse starts, open the sample cover. Look at the outlet end of the capillary in position B1.
 - If there are droplets on the outlet end of the capillary, then do step 6.
 - If there are no droplets on the outlet end of the capillary, then the capillary is blocked or the system has a pressure failure. Continue with the next step.
5. Replace the capillary, and then do the pressure rinse again.
 - If there are still no droplets on the outlet end of the capillary, then contact SCIEX Technical Support at sciex.com/request-support.
 - If there are droplets on the outlet end of the capillary, then the detection system is the only possible cause. Continue with the next step.
6. Make sure that the correct filters are installed in the LIF detector.
7. If no step change is detected, then do the calibration procedure again. Refer to the section: [Calibrate the LIF Detector \(Optional\)](#).

If the calibration procedure has been done more than 3 times, then manually set the calibration correction factor (CCF) to 1.0, and then calibrate the LIF detector again.

If the LIF detector calibration continues to fail, then contact SCIEX Technical Support at sciex.com/request-support.

Troubleshooting

6

Symptom	Possible Cause	Corrective Action
The Instrument Failure message or the System Error message is shown in the Waters Empower™ software Message Center window.	<ol style="list-style-type: none">1. The incorrect version of the GPIB driver is installed.2. The incorrect version of the .NET Language Runtime is installed.	<ol style="list-style-type: none">1. If the National Instruments GPIB driver Version 19.0 is not installed, then install it.2. If I-488.2 .NET Language Runtime 17.0.1 for .NET Framework 4.5 is not installed, then install it.
After the detector is changed, the Instrument Failure message or the System Error message is shown in the Waters Empower™ software Message Center window.	After a new detector is installed, the firmware settings were not downloaded from the PA 800 Plus system to the LAC/E module or the Instrument Server does not have the new settings.	Start the PA 800 Plus system again, and then start the LAC/E module or the computer that is physically connected to the system.
The results for the assay are very different than the results shown in the document <i>Application Guide</i> .	The parameters in the instrument method are incorrect.	<p>Examine the instrument method and make sure that:</p> <ul style="list-style-type: none">• The pressure is applied to the correct side of the capillary or both sides. Refer to the applicable document: <i>Application Guide</i>.• The pressure values are correct for the units used by the software, either millibar or psi. For instructions about how to change the pressure units used in the software, refer to the document: <i>PA 800 Plus Empower™ Driver Release Notes</i>.

Symptom	Possible Cause	Corrective Action
The results of some data processing calculations are very different than the results for similar calculations in the 32 Karat software.	Some of the capillary electrophoresis-related calculations in the Waters Empower™ software are not optimized for SCIEX systems.	Create a custom calculation for CE-specific attributes such as velocity corrected area (VCA).
Pressure or movement errors occur when the vials should be incremented during a run.	The Sample Set method is not correct.	Make sure that the vial increment number agrees with the number of lines in the sample set method and is the same as the number of runs in the sample set method.
The Scan or Channel Data Overflow error messages during data acquisition.	Because more than one PA 800 Plus system is connected to the LAC/E module, too much data is collected.	Do not acquire data on both systems at the same time. Connect each system to a separate LAC/E module.

Time Program Events

A

This section gives a list of events and the related parameters that can be added to a time program in an instrument method. Refer to the table: [Table A-1](#).

For details about the parameters, refer to the table: [Table A-2](#).

Note: The **Comment** parameter is not included in the following table but is available for every event.

Table A-1 Time Program Events

Event	Description	Parameters
Auto Zero	Zero the detector output.	At Time (min)
Capillary Temperature	Set the capillary temperature.	<ul style="list-style-type: none">• Temperature (°C)• At Time (min)
End	Identify the end of the method. Only one End event is permitted in a method. The End event must be the last event in the time program.	At Time (min)
Inject Pressure	Use pressure to inject the sample.	<ul style="list-style-type: none">• Pressure (psi or mbar)• Duration (s)• Pressure Direction• Tray Positions• Increment Every Runs
Inject Pressure Capillary Fill	Use pressure to inject the sample. This event uses a higher pressure and a longer duration than the Inject Pressure event. Use this event to completely fill the capillary with sample.	<ul style="list-style-type: none">• Pressure (psi or mbar)• Duration (s)• Pressure Direction• Tray Positions• Increment Every Runs

Table A-1 Time Program Events (continued)

Event	Description	Parameters
Inject Vacuum	Use vacuum to inject the sample.	<ul style="list-style-type: none"> • Vacuum (psi or mbar) • Duration (s) • Pressure Direction • Tray Positions • Increment Every Runs
Inject Voltage	Use voltage to inject the sample.	<ul style="list-style-type: none"> • Voltage (kV) • Polarity • Duration (s) • Tray Positions • Increment Every Runs
Lamp Off	Turn off the lamp at the specified time.	At Time (min)
Lamp On	Turn on the lamp at the specified time.	At Time (min)
Lasers Off	(LIF detector) Turn off the lasers at the specified time.	At Time (min)
Lasers On	(LIF detector) Turn on the lasers at the specified time.	At Time (min)
Relay On	Turn on the specified relays at the specified time.	<ul style="list-style-type: none"> • Relay 1 • Relay 2 • At Time (min)
Rinse Pressure	Add a Rinse event that uses pressure.	<ul style="list-style-type: none"> • Pressure (psi or mbar) • Duration (min) • Pressure Direction • Tray Positions • Increment Every Runs • At Time (min)

Time Program Events

Table A-1 Time Program Events (continued)

Event	Description	Parameters
Rinse Vacuum	Add a Rinse event that uses vacuum.	<ul style="list-style-type: none">• Vacuum (psi or mbar)• Duration (min)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)
Sample Storage Temperature	Set the temperature of the sample cooler.	<ul style="list-style-type: none">• Temperature (°C)• At Time (min)
Separate Current	Use current to separate the sample.	<ul style="list-style-type: none">• Current (μA)• Duration (min)• Ramp Time (min)• Tray Positions• Increment Every Runs• At Time (min)
Separate Current Pressure	Use current and pressure to separate the sample.	<ul style="list-style-type: none">• Current (μA)• Duration (min)• Ramp Time (min)• Pressure (psi or mbar)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)

Table A-1 Time Program Events (continued)

Event	Description	Parameters
Separate Current Vacuum	Use current and vacuum to separate the sample.	<ul style="list-style-type: none">• Current (μA)• Duration (min)• Ramp Time (min)• Vacuum (psi or mbar)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)
Separate Power	Use power to separate the sample.	<ul style="list-style-type: none">• Power (W)• Duration (min)• Ramp Time (min)• Tray Positions• Increment Every Runs• At Time (min)
Separate Power Pressure	Use power and pressure to separate the sample.	<ul style="list-style-type: none">• Power (W)• Duration (min)• Ramp Time (min)• Pressure (psi or mbar)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)

Time Program Events

Table A-1 Time Program Events (continued)

Event	Description	Parameters
Separate Power Vacuum	Use power and vacuum to separate the sample.	<ul style="list-style-type: none">• Power (W)• Duration (min)• Ramp Time (min)• Vacuum (psi or mbar)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)
Separate Pressure	Use pressure to separate the sample.	<ul style="list-style-type: none">• Pressure (psi or mbar)• Duration (min)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)
Separate Vacuum	Use vacuum to separate the sample.	<ul style="list-style-type: none">• Vacuum (psi or mbar)• Duration (min)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)

Table A-1 Time Program Events (continued)

Event	Description	Parameters
Separate Voltage	Use voltage to separate the sample.	<ul style="list-style-type: none">• Voltage (kV)• Polarity• Duration (min)• Ramp Time (min)• Tray Positions• Increment Every Runs• At Time (min)
Separate Voltage Pressure	Use voltage and pressure to separate the sample.	<ul style="list-style-type: none">• Voltage (kV)• Polarity• Duration (min)• Ramp Time (min)• Pressure (psi or mbar)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)

Time Program Events

Table A-1 Time Program Events (continued)

Event	Description	Parameters
Separate Voltage Vacuum	Use voltage and vacuum to separate the sample.	<ul style="list-style-type: none">• Voltage (kV)• Polarity• Duration (min)• Ramp Time (min)• Vacuum (psi or mbar)• Pressure Direction• Tray Positions• Increment Every Runs• At Time (min)
Stop Data	Stop data collection.	At Time (min)
Wait	Add a Wait event.	<ul style="list-style-type: none">• Duration (min)• Tray Positions• Increment Every Runs• At Time (min)
Wavelength PDA Detector	<p>(PDA detector) Change the wavelength for the specified channel in the PDA detector.</p> <hr/> <p>Note: The wavelength range (wavelength $\pm 1/2$ bandwidth) must be between 186 nm and 604 nm.</p> <hr/>	<ul style="list-style-type: none">• Channel• Wavelength (nm)• Bandwidth (nm)• At Time (min)
Wavelength UV Detector	(UV detector) Change the wavelength for Channel 1 in the UV detector.	<ul style="list-style-type: none">• Wavelength (nm)• At Time (min)

Parameters for Time Program Events

The parameters are given in alphabetic order.

Table A-2 Parameters for Time Program Events

Parameter	Details
At Time (min)	The time to start this event, given as the time from the first event with the At Time parameter equal to 0.
Bandwidth (nm)	<p>(PDA detector) The bandwidth for a Wavelength PDA Detector event, from 6 nm to 252 nm.</p> <hr/> <p>Note: The wavelength range (wavelength $\pm \frac{1}{2}$ bandwidth) must be between 186 nm and 604 nm.</p> <hr/>
Channel	(PDA detector) The channel in the PDA detector to be set to the specified wavelength.
Current (μA)	<p>The current to be applied during the event, from either $-300.0 \mu\text{A}$ to $3.0 \mu\text{A}$ or $3.0 \mu\text{A}$ to $300.0 \mu\text{A}$.</p> <ul style="list-style-type: none"> • Values from $-300.0 \mu\text{A}$ to $-3.0 \mu\text{A}$ have reverse polarity (– at the inlet and + at the outlet). • Values from $3.0 \mu\text{A}$ to $300.0 \mu\text{A}$ have normal polarity (+ at the inlet and – at the outlet).
Duration (s or min)	<p>The duration of the event.</p> <hr/> <p>Note: For pressure and vacuum events, the duration must be sufficiently long to let the system to get to the specified pressure (or vacuum). Refer to the section: About the Duration for Pressure and Vacuum Events.</p> <hr/>
Increment Every Runs	The number of runs after which the inlet and outlet vials should increment. If the vial should not increment, then type 0. Refer to the section: About Vial Incrementing .
Polarity	<p>The direction of the current to be applied during the event. Options are:</p> <ul style="list-style-type: none"> • Normal (+): + at the inlet and – at the outlet. • Reverse (-): – at the inlet and + at the outlet.

Time Program Events

Table A-2 Parameters for Time Program Events (continued)

Parameter	Details
Power (W)	<p>The power to be applied during the event, from –9.000 W to 9.000 W.</p> <ul style="list-style-type: none">• Values from –9.000 W to -0.001 W have reverse polarity (– at the inlet and + at the outlet).• Values from 0.001 W to 9.000 W have normal polarity (– at the inlet and + at the outlet).
Pressure (psi or mbar)	<p>The pressure to be applied during the event.</p> <hr/> <p>Note: The system requires time to get to the specified pressure. If the Duration parameter is too short, then the system cannot get to the specified pressure. Refer to the section: About the Duration for Pressure and Vacuum Events.</p> <hr/>
Pressure Direction	<p>The direction of the pressure to be applied during the event. Options are:</p> <ul style="list-style-type: none">• Forward: From inlet to outlet.• Reverse: From outlet to inlet.• Simultaneous: In both directions at the same time.
Ramp Time (min)	<p>The time required for the system to get to the specified pressure, voltage, power, or current.</p>
Relay 1	<p>The relay to be opened or closed.</p>
Relay 2	<p>The relay to be opened or closed.</p>
Temperature (°C)	<p>The temperature for the cartridge or sample cooler.</p>
Tray Positions	<p>The inlet and outlet vials for the event. For each vial, identify a tray and a position. Refer to the section: About Tray Positions.</p>
Vacuum (psi or mbar)	<p>The vacuum to be applied during the event, from 0.1 psi to 5.0 psi (or 6.9 mbar to 344.7 mbar).</p> <hr/> <p>Note: The system requires time to get to the specified vacuum. If the Duration parameter is too short, then the system cannot get to the specified vacuum. Refer to the section: About the Duration for Pressure and Vacuum Events.</p> <hr/>

Table A-2 Parameters for Time Program Events (continued)

Parameter	Details
Voltage (kV)	The voltage to be applied during the event, from –30.0 kV to 30 kV for any of the Separation Voltage events and –10.0 kV to 10 kV for the Inject Voltage event. The Polarity parameter sets the direction of the voltage.
Wavelength (nm)	The wavelength for the event, from 190 nm to 600 nm.

About the Duration for Pressure and Vacuum Events

The system requires time to get to the specified pressure (or vacuum). If the duration is too short, then the system cannot get to the specified pressure or vacuum. Use the following tables to make sure that the duration is sufficiently long.

Table A-3 Required Duration to Get to the Specified Pressure

To get to this pressure...		Set the duration to at least...
0.1 psi	6.9 mbar	1.0 sec
0.2 psi	13.8 mbar	1.5 sec
0.3 psi	20.7 mbar	2.0 sec
0.4 psi	27.6 mbar	2.5 sec
0.5 psi	34.5 mbar	3.0 sec
0.7 psi	48.3 mbar	3.4 sec
2.0 psi	137.9 mbar	3.5 sec
5.0 psi	344.7 mbar	3.8 sec
9.5 psi	655.0 mbar	5.0 sec
25.0 psi	1723.7 mbar	6.3 sec

Table A-4 Required Duration to Get to the Specified Vacuum

To get to this vacuum...		Set the duration to at least...
0.10 psi	6.9 mbar	2.0 sec
0.15 psi	10.3 mbar	2.5 sec
0.30 psi	20.7 mbar	3.0 sec
0.40 psi	27.6 mbar	3.5 sec

Table A-4 Required Duration to Get to the Specified Vacuum (continued)

To get to this vacuum...		Set the duration to at least...
0.50 psi	34.5 mbar	4.0 sec

About Tray Positions

The following **Tray Positions** parameters are used to identify the positions of the capillary inlet and outlet for Rinse, Inject, Separate, and Wait events:

- **Inlet Vial:** The inlet vial for the next event, from A1 to F6.
- **Inlet Tray:** The inlet tray for the next event, either **Buffer** or **Sample**. For Inject events, **Sample List** is also available. Refer to the section: [Sample Vial Positions for Inject Events](#).
- **Outlet Vial:** The outlet vial for the next event, from A1 to F6.
- **Outlet Tray:** The outlet tray for the next event, either **Buffer** or **Sample**. For Inject events, **Sample List** is also available. Refer to the section: [Sample Vial Positions for Inject Events](#).

In the PA 800 Plus system, the geometry of the sample and buffer trays and the dimensions of the capillary cartridge control the access to all 36 positions in the tray. For example, if the capillary inlet is in position A6 in the buffer inlet tray, then the capillary outlet cannot get access to position F6 in the buffer outlet tray. These incompatible positions are referred to as *tray collisions* or *vial collisions*.

The software makes sure that the tray positions are satisfactory. If a collision is possible, then a warning message is shown.

The following table shows column combinations that do not cause a collision.

Table A-5 Inlet and Outlet Column Combinations That Do Not Cause a Collision

Inlet Columns	Compatible Outlet Columns
A to F	A to C
B to F	A to D
C to F	A to E
D to F	A to F

Sample Vial Positions for Inject Events

The **Inject** event is used to inject the sample into the capillary before the separation starts. The positions of the vials that contain the sample for **Inject** events can be specified in the instrument method or in the sample set method.

1. To set the vial positions in the instrument method, edit the **Tray Positions** parameter for any **Inject** event.
2. To set the vial positions in the sample set method, do the following:
 - a. In the instrument method, select **Sample List** for the **Inlet Tray** in the **Tray Positions** parameter.
 - b. In the sample set method, edit the vial positions in the **Plate/Well** field.

About Vial Incrementing

Vial incrementing is an automated process in which the system moves the inlet and outlet vials forward after a specified number of method cycles. Vial incrementing removes the need to create new methods when different vial positions are required during a set of instrument runs. One method can be used to change to new vial positions automatically.

If vial incrementing is not used, then vials might overflow with liquid that collects in the interface block, pressure manifold, and other parts of the system. Vial incrementing also helps to prevent the depletion of the ionic strength of the buffer.

Vial incrementing can be set for either or both of the inlet and outlet vials.

To use vial incrementing, type a value for number of runs in the **Inlet** and **Outlet** fields of the **Increment Every Runs** parameter. Runs are the number of times a method set is done before vial incrementing occurs.

When the sample set method advances to a new method set, vial incrementing starts again.

Plate Definition Files

B

This section includes the plate definitions for the buffer tray, sample tray, and SCIEX 96-well sample tray. These plate definitions must be set in the Waters Empower™ software.

The definition files should be installed as part of the PA 800 Plus Empower™ Driver installation. If the definition files are missing and the plates require a definition, then copy the text, paste it in a text editor, and then save the file.

PA800Plus Sample Tray Plate Definition File

```
Empower Profile for Plate Type: CE Sample Tray
Plate Type: XY
Permanent: No
Plate Terminology: Plate
Well Terminology: Well
Plate Dimensions:
  X: 85.00
  Y: 128.00
  Height: 17.00
Well Dimensions:
  Top Left Well X Location: 9.00
  Top Left Well Y Location: 17.10
  Well Diameter: 12.00
  Well Depth: 14.00
Row and Column Dimensions:
  Number of Rows: 8
  Row Spacing: 13.40 mm
  Number of Columns: 6
  Column Spacing: 13.40 mm

Row and Column Offsets:
  Row Offset Type: None
  Row Offset: 0.00 mm
  ColumnOffset Type: None
  Column Offset: 0.00 mm
Origin: Bottom Left

Scheme:
  Referencing: XY
  Horizontal: ABC ...
  Vertical: 123 ...
  Sequential Continuous: Off
  Horizontal First Priority: On
```

PA800Plus 96 Well Sample Tray Plate Definition File

Empower Profile for Plate Type: 96-Well Sample Tray

Plate Type: XY

Permanent: No

Plate Terminology: Plate

Well Terminology: Well

Plate Dimensions:

X: 85.00

Y: 128.00

Height: 17.00

Well Dimensions:

Top Left Well X Location: 11.00

Top Left Well Y Location: 14.50

Well Diameter: 6.80

Well Depth: 14.00

Row and Column Dimensions:

Number of Rows: 12

Row Spacing: 9.00 mm

Number of Columns: 8

Column Spacing: 9.00 mm

Row and Column Offsets:

Row Offset Type: None

Row Offset: 0.00 mm

ColumnOffset Type: None

Column Offset: 0.00 mm

Origin: Bottom Left

Scheme:

Referencing: XY

Horizontal: ABC ...

Vertical: 123 ...

Sequential Continuous: Off

Horizontal First Priority: On

PA800Plus Buffer Tray Plate Definition File

Empower Profile for Plate Type: CE Buffer Tray

Plate Type: XY

Permanent: No

Plate Terminology: Plate

Well Terminology: Well

Plate Dimensions:

X: 85.00

Y: 85.00

Height: 17.00

Well Dimensions:

Top Left Well X Location: 9.00

Top Left Well Y Location: 9.00

Well Diameter: 12.00

Well Depth: 14.00

Row and Column Dimensions:

Number of Rows: 6

Row Spacing: 13.40 mm

Number of Columns: 6

Column Spacing: 13.40 mm

Row and Column Offsets:

Row Offset Type: None

Row Offset: 0.00 mm

ColumnOffset Type: None

Column Offset: 0.00 mm

Origin: Bottom Left

Scheme:

Referencing: XY

Horizontal: ABC ...

Vertical: 123 ...

Sequential Continuous: Off

Horizontal First Priority: On

Topics for Familiarization

C

During installation, the SCIEX field service employee (FSE) should have familiarized or reviewed the following procedures with the customer:

- Software functions:
 - How to install the USB license key
 - How to create, edit, and save instrument methods
 - How to configure the software to use multiple plates
 - How to use Direct Control and the following components:
 - Instrument status
 - Status field
 - Parameter tabs and buttons
 - How to run a single sample or a sample set method
 - How to stop a run
- How to view error messages in the Waters Empower™ software Message Center window
- How to install a cartridge
- How to load samples
- For systems with more than one detector, how to change the detectors
- Maintenance procedures

Contact Us

Customer Training

- In North America: NA.CustomerTraining@sciex.com
- In Europe: Europe.CustomerTraining@sciex.com
- Outside the EU and North America, visit sciex.com/education for contact information.

Online Learning Center

- [SCIEX Now Learning Hub](#)

Purchase Supplies and Reagents

Reorder SCIEX supplies and reagents online at store.sciex.com. To set up an order, use the account number, found on the quote, order confirmation, or shipping documents. Currently, customers in the United States, Canada, United Kingdom, Belgium, Netherlands, France, Germany, and Switzerland have access to the online store, but access will be extended to other countries in the future. For customers in other countries, contact a local SCIEX representative.

SCIEX Support

SCIEX and its representatives maintain a staff of fully-trained service and technical specialists located throughout the world. They can answer questions about the system or any technical issues that might arise. For more information, visit the SCIEX website at sciex.com or contact us in one of the following ways:

- sciex.com/contact-us
- sciex.com/request-support

Cybersecurity

For the latest guidance on cybersecurity for SCIEX products, visit sciex.com/productsecurity.

Documentation

This version of the document supersedes all previous versions of this document.

To find software product documentation, refer to the release notes or software installation guide that comes with the software.

To find hardware product documentation, refer to the documentation that comes with the system or component.

The latest versions of the documentation are available on the SCIEX website, at sciex.com/customer-documents.

Note: To request a free, printed version of this document, contact sciex.com/contact-us.
