

Using the Fraction Collector in Empower 3 Environment

Technical Note

Technical Guide for the configuration and use of the Agilent Fraction Collectors with Waters Empower

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Introduction

When using an Agilent LC without an automated sampler, a manual injector is required to perform a run, e.g. G1328C Agilent 1260 Infinity II Manual Injector.

Waters Corporation's adoption of the Agilent Instrument Control Framework (ICF) for their Empower Data System is called *Agilent ICF Support*. The *Waters ICF Support* is part of the Waters Instrument Control Package (ICS).

This guide describes how to configure Agilent Fraction Collectors in a Waters Empower environment.

Waters Instrument Control Packages	Agilent ICF Agilent LC Driver	Fraction Collection support in Waters Empower 3
ICF Support v3.2 With ICF 2.6 Update 2 # 667006157	A.02.06 U2 A.02.19 SR2	supported
ICF Support v3.2 #667006057	A.02.05 A.02.18 supported	
ICF Support v3.1 #667005859	A.02.05 A.02.18	
ICF Support v3.0 With ICF A.02.5 Update #667005815	A.02.05 A.02.18	
ICF Support v3.0 #667005586	A.02.04 A.02.14	
ICF Support v2.2 With ICF A.02.04 Update #667005584	A.02.04 A.02.14	Supported
ICF Support v2.2 #667005449	A.02.03 DU2 A.02.13	
ICF Support v2.1 HF1 #667005397	A.02.03 DU1 HF2 A.02.11 SP1 HF2	
ICF support v3.0	A.02.04 A.02.14	
ICF Support v2.1 HF1 #667004920	A.01.05 SP1	
ICF Support v2.1 HF1 #667004900	A.01.04	Not supported
and any lower integration	and any lower integration	

Table 1 Supported and unsupported configurations

Table 2 Supported Agilent Fraction Collectors

Product No.	Module Name	Min. FW
G1364A	1100 Automatic Fraction Collector	A.06.53
G1364B	1260 Infinity Preparative-scale Fraction Collector	A.06.53
G1364C	1260 Infinity Analytical-scale Fraction Collector	A.06.53
G1364D	1260 Infinity Micro-scale Fraction Collector	A.06.53
G5664A	1260 Infinity Bio-inert Fraction Collector	A.06.53

Adding the Fraction Collector to the Agilent LC Instrument System

NOTE Ensure that all Agilent LC modules in the LC system meet or exceed the minimum firmware requirements specified by the 3rd-party CDS software vendor and meet Agilent's firmware set/firmware interoperability requirements. Agilent proposes to use the latest available firmware set.

https://www.agilent.com/en-us/firmwareDownload?whid=69761

Adding the Fraction Collector to the Agilent LC Instrument System

First prepare the instrument set up:

- 1 Close the Empower CDS.
- 2 Switch off the Agilent LC Tower and add the Agilent Fraction Collector to the LC System.
- **3** Connect the power plug and use the CAN cable to connect the fraction collector module with one module of the existing Agilent LC system. If additional information is required, please consult the user manual of the fraction collector in question.
- 4 Switch on all modules in the Agilent LC instrument system.
- 5 Put the required vial tray, wellplate tray, or tube tray in the Fraction Collector.
- 6 If wellplates are in use: Place all required wellplates in the Fraction Collector.
- 7 Close the Fraction Collector door.

Configuration of the Fraction Collector

Configure Fraction Collector in Empower

The configuration of the Fraction Collector in Empower requires the PreConfiguration Utility.

Agilent provides the **PreConfiguration Utility** as part of the ICF installation, starting with ICF A.02.01, available in Empower since ICF Support v 2.1 HF1 / ICF A.02.03 DU1 HF2.

The **PreConfiguration Utility** can be found at one of two locations:

- ICF Support v 2.1 HF1: Using Empower installation on the LAC/E box (see "Using Empower Installation on the LAC/E box" on page 3).
- ICF Support v 2.2 and higher: Via the Empower Configuration Manager (recommended for Client/Server configuration) (see "Via the Empower Configuration Manager" on page 4).

Using Empower Installation on the LAC/E box

1 Navigate to <drive letter>:**\Empower\Instruments\AgilentLC** and double-click the PreConfiguration Utility.

ICF Agilent.InstrumentControl.InstrumentPreConfigurator.exe

Figure 1 PreConfiguration Utility

Configuration of the Fraction Collector

Via the Empower Configuration Manager

Software required: ICF Support v2.2 or higher (See "Known Issues and Limitations" on page 16)

1 In the Empower Configuration Manager, select Tools > Agilent PreConfiguration.



Figure 2 Configuration Manager

2 Enter the IP address or host name of the LAC/E box that your instrument is connected to into the pop-up screen **Configuration Directory** and click **Connect**.

NOTE

Do not enter the IP address of the instrument here.

📁 Configuration Direct	ory: Connected to 16	9.25 💼 🔳 💌
IP Address / Host Name		Connect
Del	ete Configure	Exit

Figure 3 IP address to connect

3 Once the IP address is connected, click New to open the PreConfiguration Utility.

Regardless of which PreConfiguration Utility was chosen, continue the configuration using the following steps when InstrumentConfigurations opens:

1 Configure the Agilent system using **AutoConfigure**. All online LC modules (connected via CAN cables) are automatically detected and added into the right-hand window.

Aglert ELSD 38×ELSD 1280 ELSD 1280 ELSD 1290 ELSD 0.01200/1260/1290 LC 0.4 Aglert 1100/12001/200 LSystems 0.4 Aglert 7100 CE		Bin: Pump (G1312A,DE53500134) Sampler (G1329A,DE80301699) DAD (G1315B,DE11112497) Fraction Collector (G1364B;DE40501077)	
	Auto Configure		
Heb		Up Down Configure DK	E Clear

Figure 4 Fraction Collector configuration set up

Configuration of the Fraction Collector

2 Double-click on the Fraction Collector in the left side of the window or select it and click **Configure** at the bottom of the screen.

Communication					Options	
1	Device name	Fraction	n Collector			
	Type ID	G13648	•			External contacts board installed
:	Serial number	DEACS	00460			Thermostat installed
Firm	ware revision	A.06.53	3 [005]			MSD installed
		Conne	ection settings			
Peak detectors					Inner Capillary	
Detector	Serial					0.0
G1315C	DE52500	247		Ŷ		u.emm • mm
				¥		
					Needle Type	
						Preparative •
Add			Remove			
				Define W	/ellplates	

The Fraction Collector configuration window opens.

Figure 5 Configuration Screen of the Fraction Collector

3 In the **Configure Fraction Collector** dialog, the following set points/information can be defined:

Communication: This information is automatically determined when first connecting with the module.

Peak Detector: Present detectors in this screen can be used as peak triggers for the Fraction Collector. They are detected during auto-configuration. Up to 4 peak detectors are supported.

- **a** Define the required peak detector.
- **b** If you have several peak detectors, specify the order by using the up and down arrows.

Options: Additional options for the Fraction Collector can be selected. Additional options are graphically indicated in the LC status window (e.g. **Thermostat installed**, **External contact board installed**) and enable additional method settings for these options.

NOTE In Empower environment mass-based fraction collection is not supported. Regardless, the option becomes available as soon as a G1390A Universal Interface Box (UIB) is connected.

Inner Capillary: Specify the inner capillary to be used. The value needs to be set according to the capillary in use. This parameter is not detected automatically.

Needle Type: Depending on the physically built-in needle, the needle type must be set manually. This parameter is not detected automatically.

Define Wellplates...: This dialog offers various predefined wellplates currently available, as well as the possibility to add customized plates. You cannot assign which plates will be used for a run in this dialog. This can only be done in the LC Status Dashboard.

NOTE

The wellplates used for the Fraction Collector need to be set here and in **Empower > Configuration Manager > Plate Types**. Ensure that the sample to be used is defined in both dialogs.

Note that wellplate changes can be triggered by a user changing the tray or a plate configuration on purpose, but a change is also triggered by the system when

- a tray is not in the fraction collector during startup of the software
- the fraction collector door is open while software starts up
- a tray is changed during the runtime of the fraction collector a change on the fly is not possible.

Before starting **Run Sample** in Empower ensure that

- a tray is present in the fraction collector. In case of a wellplate tray, the plates are already placed on the tray.
- the fraction collector door is closed.

Start Empower **Run Sample** screen. The LC Status window automatically displays all available online modules. The Fraction Collector shows the identified tray (vial tray, tube tray or wellplate tray).

Figure 6 exemplarily shows a recognized tray example of a new instrument with a 4 plate wellplate tray.

Fraction Co	I ? <u> </u>	
()on ⊜off	ldle EMF⊘	
↓ • n/≥		

Figure 6 New instrument with a 4 plate wellplate tray

NOTE

NOTE

After this first instrument start up not all configuration items are defined. For example, you need to define the wellplates you want to use or add the tube volume and height information if you want to use a tube tray.

If the instrument was never used with this tray before, the tray is recognized, and the last known plates are used. You need to assign the correct plates currently present in the next step.

If the instrument was already used before, the last known plate configuration for this tray used by this instrument appears.

If the user requires a different plate type and changes them in the tray, a new assignment is required. Figure 7 shows a recognized tray example of an instrument with a 2 plate/10 vial tray, which was previously used on this instrument with those assigned plates.



Figure 7 Instrument with previously assigned plates

Assign Wellplates or Tube Information

There are two ways to assign these additional configurations

- via Module Option (recommended) OR
- via right-clicking on the Fraction Collector tile

Agilent strongly recommends using the Module options to change the configuration settings.

The Fraction Collector offers additional configuration parameters, e.g. the fraction order.

1 To define these parameters via the **Module Options**, select the corresponding tab below the LC Status Dashboard.



Figure 8 Agilent LC Instrument Status Window

2 Click Module Options.

NOTE

Select the Module Options	button in order to configure runtime instrument parameters.	
Module Options		

Figure 9 Module options button

3 Define the parameters in the Soft Configuration.

Tray A/B/C – the tray is automatically recognized during the software startup. If the tray is *not* recognized, verify that there is no hardware related issue.

- The tag is not written correctly (try another tray)
- A defect antenna (try the same tray in another fraction collector)

8

Plate Type (not accessible when using tube trays)

The LC driver offers these plate types, for example 384 Agilent.

AgilentLC#AFRACO Soft Configuration							×
Tray Configuration							
Tray A: 2P+10Vials Wellplates	Tray B:	No t	ray		Tray C: No	tray	
Plate Type (No Plate>				•	Installed	back left	back right front right
Collection Order					Collection	n Mode	
					C	Discrete fractions	
Row by row		0	Shortest path by co	lumn	C) Continous flow	
					Additiona	I Fraction Locations	
Column by column		0	Shortest path by row	4		None	•
Reserved Locations				Peak Detector Del	lay Volumes		
				Detector	Serial	Delay Volume [µl]	
				G1315C	DE52500247	0	
Row(s)							
Column(s)							
Single Location(s)							
Needle Movement				Linked Pump			
				G1312	2A:DE53500134	•	
				T			
Above location di	stance		0,0 📫 mm	Tube			
					Tube Volu	ime 0,0	10 0 mL
					Tube Hei	ght	0 📜 mm
			OK	Cancel			

Figure 10 Fraction Collector Options Wellplate and Vial Tray

Collection order

Offers the possibility to define the collection order strategy, for example **Row by row**. Detailed information is provided in the online help. Press **F1** while in the Soft Configuration window.

Collection mode

Offers the definition how the collection takes place

- **Discrete fractions**: The diverter valve switches to waste before moving on.
- Continuous flow: The flow continues while moving to the next position.

Detailed information is provided in the online help. Press **F1** while in the Soft Configuration window.

Additional Fraction Locations (Not supported in Empower environment)

Offers additional fraction collection locations like 10 vials or funnel bars. This option is not supported in Empower Environment.

Reserved Location

Allows to specify locations that will not be used for fraction collection. Detailed information is provided in the online help. Press **F1** while in the Soft Configuration window.

Peak Detector Delay Volume

Use Lab Advisor for the delay volume calibration. The delay volume is written to the Fraction Collector module and read out by the Driver. It is also possible to manually enter the delay volume.

Needle Movement

Allows to specify how the need moved into/above the fraction collection. Detailed information is provided in the online help, Press **F1** while in the Soft Configuration window.

Linked Pump

If required, this parameter needs to be set manually. This function should be used, as the delay volume calibration will enable this volume to be calculated based on new flow rates set on the pump.

Tube (parameter only active if a tube tray is recognized)

These parameters are required when using tubes and are automatically filled by the system for Agilent consumables.

Andenti C#AFRE DI Soft Configuration			
Tay Configuration			
Trav A: 126 Tubes	Tray B: No tray		Tray C: No tray
Wellplates	,,		
Plate Type (No Plate) treat as 4 x 96		Ŧ	back left back right Installed at front left front right
Collection Order			Collection Mode
			 Discrete fractions
	O Shortest path by	column	O Continous flow
**** _			Additional Fraction Locations
Column by column	Shortest path by i	ow	None 👻
			·
Reserved Locations		Peak Detector Del	ay Volumes
		Detector	Serial Delay Volume [µ]
		G1315C	DE52500247 0
Row(s)			
Column(s)			
Single Location(s)			
origio zoodion(o)			
Veedle Movement		Linked Pump	
todae movement		Cancer any	
		G1312	2A:DE53500134 👻
		Tube	
Above location dista	ance 0,0 🕻 mm	Tube	Tube Volume 12,00 ; mL Tube Height 100 ; mm
	OK	Cancel	

Figure 11 Fraction Collector Options Tube tray

NOTE

Important: Any change in this dialog requires a reboot and a new auto-configuration using the **PreConfiguration Utility.**

Additional Fraction Collector Configuration via Fraction Tile

1 Right-click on the Fraction Collector Tile. Select Assign Wellplates....



Figure 12 Assigning wellplates via the LC status window

2 In the Tray and Plate Configuration dialog, select the plate type you want to use.

😁 Tray and Plate Configura	tion			- • ×
Tray Configuration				
Tray A: 2P+10Vials	Tray	B: Notray	Tray C: No tray	
Plate Configuration				
Plate Type	<no plate=""> <no plate=""> "35VialRack" "40viaPlate" "54ViaPlate" "54ViaPlate" "15HRV5mViaPlate" "15HRV5mViaPlate" "5HRV5mViaPlate" "5HRV5mViaPlate"" "5HRV5mViaPlate" "5HRV5mViaPlate" "5HRV5mViaPlate" "5HRV5mViaPlate"" "5HRV5mViaPlate" "5HRV5mViaPlate" "5HRV5mViaPlate" "5HRV5mViaPlate"" "5HRV5mViaPlate""" "5HRV5mViaPlate"" "5HRV5mViaPlate""" "5HRV5mViaPlate""""""""""""""""""""""""""""""""""""</no></no>			- A

Figure 13 Plate Type selection

NOTE

Important: Any change in this dialog requires a reboot and a new auto-configuration using the **PreConfiguration Utility**.

The Fraction Collector method

- 1 If the currently loaded configuration is not in synch with the loaded **Instrument Method**, the system requests the user to update the given method configuration with the new instrument configuration. Click **OK** and the system opens the method dialog.
- 2 Set the parameters:

Instrument Configuration tab: In the **Instrument Configuration** tab, the LC system configuration is available. Changes in this section of the method are not applied to the system. All settings in this dialog, as well as any changes such as modifying the needle type must be set in the PreConfiguration tool.

🐴 Untitled in test as System/Administrator - Instrument Method Editor	00 8
Internet Noted Present Noted Audus Davreb Gread Internet Collegation () Advert 1500 () A	Bite Prove Bit 124-04252000144 Hit Bis service (Bit 137:054000087HB) Disk (Bit 157:054000087HB) Therefore CodeNet (Bit 1444 Bite Science (Bit 1444 Bite Science (Bite Bite Bite Science (Bite Bite Bite Science (Bite Bite Science (Bite Bite Bite Science (Bite Bite Bite Science (Bite Bite Bite Bite Bite Science (Bite Bite Bite Bite Bite Bite Bite Bite
	Up Down Configure Clear

Figure 14 Fraction Collector Configuration Screen in instrument method

General Tab: Select the Start at Location check box and provide the fraction start location.

The syntax of the fraction start location is important. Only correct locations parameters enable a start of the run/sequence. Ensure to follow the schema, for example: P1-A-1.

Instrument Method Pretreatment Method Auxiliary Channels	General	Instrument Configuration
Fraction Collector Options Image: Start at Location: Image: P1-A-1		

Figure 15 Fraction Collector Options

Instrument Method tab: The instrument method tab provides access to all method parameters of the LC system, one tab per module. Select the **Fraction Collector** tab to enter the method parameters.

ntitled in test Edit View	as Sys Hele	item/Administr	rator - Instrur	nent Meth	od Editor							(- (
	×												
nent Method	Pretr	reatment Method	i Auxiliary Ch	annels Ge	nesal Instrume	nt Configuration							
ry Pump Hi	iP Sang	pler DAD Fr	raction Collect	я									_
										Fra	ction Collector (G1364B)	
action Trigge	er Mode)					🖌 Timetable (empty)					
۲	Qft						Time (min)	Function	Parameter				
0	Peak	based	max peak di	ration		min							
0	Time	-based with num	ber of fraction										
0	Time	-based with times	sices			min							
ak Detector				0									
etector	Unit	Mode	Slop	e Skope	Threshold	Upper Threshold							
1315C.DE	mAIJ	OII	5.00	5.00	5.000	3000.000							
2500247													
	Frac	tion is collected	when a peak	s detected I	by								
	0	all peak detecto	242										
	۲	at least one pea	sk detector										
optime			Po	time									
At F	umodr	niector		۲	Off		Add	Berrove	Clear all				
õ 🗆		1.00 0 min		õ		1 min	Cut	Corv	Pate				
								- 097					
							Advanced						

Figure 16 Fraction Collector Method Screen

Left Hand side:

Fraction Trigger Mode:

Off (no fraction triggering)

Peak-based (at least one peak detector must be set)

Time-based with number of fractions (ensure to enter a stop time)

Time-based with time slices

Mass-Based fraction collection requires the option **MSD installed** to be enabled. When using an UIB to connect to an MS, this setting is available. Keep in mind that the combination of Fraction Collection and MSD is not supported.

For the delay volume calibration use Lab Advisor. The delay volume is written to the Fraction collector module and read out by the Driver. Also, it is possible to manually enter the delay volume.

Peak Detector parameters

The detector being used to trigger the Fraction Collector is listed along with peak detector parameters for slopes and threshold.

Right Hand Side:

- Timetable
- Advanced set points

Rinse Fraction Collector Needle (Default: no selection)

Auxiliary - Max. fill volume per location (Default: as configured)

Changing Trays and Plates using the Fraction Collector

Changing a tray and changing plates in the Fraction Collector is a change of the configuration.

The instrument and software need to adjust the parameters accordingly. Examples are the number of locations, the tube height/volume, the position numbering. These changes have to be written to the instrument and read out by the software as the system needs to be synchronized. In Third Party environment, this synchronization requires a restart of the instrument control PC or, if PreConfiguration is used, a new auto-configuration.

A configuration change is triggered

- if no tray is present in the fraction collector during software start-up.
- if the fraction collector door is open during software startup.
- if the tray is exchanged during run time.
- if the assigned wellplate is changed.

Example:

How to change from a 2 plate/10 vial tray to a wellplate tray with 4x 384 wellplates:

1 The current set up shows the 2 plate/10 vial tray. Changing to the 4x 384 wellplates requires a tray change.

😫 Tray and Plate Configura	tion		
Tray Configuration			
Tray A: 2P+10Vials	Tray B: No tray	Tray C: No tray	
Plate Configuration			
Plate Type	*96Greiner*		-
	Treat 384 as 4x96	back left 💟 🗌 back right	
		front left 🔽 📃 front right	
		Ok Cancel	Help

Figure 17 Tray and Plate Configuration dialog

- 2 Exchange the tray in the fraction collector. Place the 384 wellplates on the tray.
- 3 Close the fraction collector door and wait until the system finishes the arm movement.

The LC Status dashboard of the fraction collector still shows the 2 plates/10 vial tray, the sequence locations offer less location, etc. as the new configuration is not synchronized between instrument and software.

😫 Tray and Plate Configuration					
- Tray Configuration					
Tray A: 2P+10Vials					
Plate Configuration					
Plate Type *96Greiner*					

Figure 18 Tray Configuration

- 4 Close Run Samples and open the Empower Configuration Manager.
- 5 Reboot the LAC/E.
- 6 Start the **PreConfiguration Utility**.
- 7 Delete the existing instrument and perform a new auto-configuration.
- 8 Close the PreConfiguration Utility.

- **9** Start **Run Samples**. The LC Dashboard Fraction collector tile shows the updated tray information. The last assigned plates are displayed in the graphical interface:
 - If this tray has not been on this system before, the plates' locations are empty.
 - If this tray was previously used with 4x 384 plates, the plates are already correctly assigned as the instrument stored this information.
 - If the tray was used with another plate, that plate assignment will appear in the graphical interface.

Fraction Coll.	
	Idle
() On COff	EMF⊘
۲ • • • • • • • • • • • • • • • • • • •	1

Figure 19 New wellplate tray recognized

Tray and Plate Configura	tion	
Tray A: 4 Plates	Tray B: No tray	Tray C: Notray
Plate Configuration		
Plate Type	<no plate=""></no>	
	Treat 384 as 4x96 Installed at	back left back right front left front right
		Ok Cancel Help

Figure 20 Tray after change recognized, no plates defined after recognition

10 Assign the plate type either via the Module options or the LC Status Dashboard Fraction Collector tile. To do the latter, right-click and select **Assign wellplates > Assign plates to the tray**. Changing the plate here, the tile will briefly go to status yellow as it writes the new configuration to the instrument.

		I	"3845.reiner" "384GreinerSmallVolume" "24WhatmanUniPlateU"				
-	Comula Matulu	Function	"96Corning" "96CorningV" "284Corning"				
_	Sample matrix	rancaon	*96EppendorfC*				
-			*96M.IBesearchC*				
Ē			*384MJResearchC*				-
ſ	😭 Tray and Plat	te Configura	"96ApogentC" "384ApogentC"				
I	– Tray Configurati	on	*96AgilentM*				
I			*96MicromassSampleM*				
I	Tray A:	4 Plates	*96MicromassLockM*				
I			*84MicromassSampleM*				
I	- Plate Config	uration	"84MicromassLockM"			· ·	
		Dista Tura	dia Diata			_	
		mate i ype	<no plate=""></no>			•	
			Trest 204 as 4v90	Installed at	back left 📃 📄 back right		
1			11eat 304 as 4x30	inisidileu al	front left 📃 📄 front right		
1							
					Ok Cancel	Help	

Figure 21 Plate Configuration

- **a** Close the CDS and reboot the LAC/E.
- **b** Perform a new auto-configuration using the PreConfiguration Utility.
- c Restart the run samples.

Fraction Coll ? _ 🗆								
	Idle							
() On COff	EMF⊘							
	* n/s							
384Agilent *384Agilent*								
Figure 22 Wellplate tray								

Known Issues and Limitations

Following functions /features are not supported in Empower Environment:

- MS installed
- Additional Fraction Locations
- The 10 vial position on a **10 vial position on a 2Plate/10 vial tray** is not supported

Appendix

Waters TechNotes

Additional information on the PreConfiguration Utility is provided on the Waters Support Webpage:

http://www.waters.com/waters/support.htm?lid=134936402&cid=511442&type=TECN

Document reference: TECN134936402

Title: Using the Agilent PreConfiguration Utility with Agilent ICF Support Version 2.2

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