

RapidFire 365 High-throughput Mass Spectrometry System

Quick Start Guide

For Research Use Only. Not for use in diagnostic procedures.

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What is the RapidFire 365 High-throughput Mass Spectrometry System?

The RapidFire 365 High-throughput Mass Spectrometry System helps to eliminate bottlenecks in your drug discovery workflow and enhance the screening process of sample analyses. By feeding samples directly to the mass spectrometer, RapidFire technology enables the acceleration of drug discovery pipeline decisions.

The high-throughput mass spectrometry system consists of a proprietary sample purification and injection system that interfaces directly with the mass spectrometer. The RapidFire-MS system can simultaneously analyze multiple analytes in complex mixtures presented by biological samples and remove contaminants such as salts, buffers, detergents, proteins, microsomes, and subcellular material.

The RapidFire system facilitates the analysis of one 384-well plate in about 50 minutes or as many as ten 384-well plates (3840 individual samples) in a single instrument shift (8 hours). When you run an experiment continuously over a weekend, the system can run 63 plates (24192 individual samples). The throughput of the RapidFire system for label-free biologically-relevant data approaches that of optical technologies.

The system is equipped with one BenchBot sample handler and four plate stacks. The plate stacks enable the system to process 63 microplates in one batch. Three plate stacks serve as the input stacks, and each can accommodate 21 microplates. One plate stack is left empty to serve as the output stack. When all of the plates from one input stack is moved to the output stack, the original input stack becomes the output stack. In addition, the bar code scanner is capable of reading bar code labels on any side of the sample plates.



Figure 1. BenchBot sample handler and input stacks.

What's New in RapidFire Integrator 5.0

- Added support for:
 - MassHunter Data Acquisition for Ultivo LC/TQ C.01.00
 - MassHunter Data Acquisition for TOF/Q-TOF LC/MS B.09.00
 - MassHunter Data Acquisition for Triple Quad LC/MS B.09.00

What's New in RapidFire Integrator 4.3

- Added Support for Windows 10 (in addition to Windows 7)
- Support for these Agilent MassHunter Data Acquisition revisions:
 - MassHunter Data Acquisition for TOF/Q-TOF LC/MS B.08.00
 - MassHunter Data Acquisition for Triple Quadrupole LC/MS B.08.02
- Added support for RoHS-compliant G9531A RapidFire 365 High-throughput Mass Spectrometry system
- Sciex LC/MS instruments no longer supported

Where to find more information

User Guides



You can access these guides from the RapidFire Resource App which you can start from the RapidFire Resources icon.

User Guide This guide contains information to prepare, maintain, and run the RapidFire system.

Data Analysis Guide Use this guide to learn how to use RapidFire Integrator to prepare and analyze data.

Safety Guide This guide contains safety instructions for operating the RapidFire system.

Web Site

Go to www.agilent.com for more information about RapidFire.

Getting Started

Refer to the User Guide for detailed instructions.

System Preparation

Steps		Details				
1	Prepare solvents	Pull out the solvent drawer and make sure you have enough solvent.				
2	Check waste capacity	Check that the waste flasks have enough room to accommodate the waste that will be generated from the runs that are planned.				
3	Start the vacuum system	Open up the vacuum valve or switch on your stand-alone vacuum pump.				

System Preparation

Steps	Details						
4 Start the software	 Double-click the RapidFire Control Panel desktop icon. Click File > Choose CFGs. Select the configuration folder under C:\Agilent\ RapidFire. Click Start in the Control Panel dialog box. 						
	RapidFire Control P — — X File						
	RapidFire Wash Station Pump 1 Sipper						
	Pump 2 Sample Interface Pump 3 Column Changer Pump 4 Plate Handler MC 1						
	MC 2 MassHunter Client LogServer RFRemServer						
5 Verify network settings	Stop Stor 1 Double-click the RapidFire UI desktop icon.						
	 Click System Tools > Network Settings. Verify that the IP addresses for the RapidFire Integrator computer and the mass spectrometer computer are correct. Click Connect in the main window. 						
	RapidFire: cfgs(4.0.X)_#RF365 File Batch Configuration Tools Plateset Run Controls Network Settings Plates Plates Plates Mode Settings Sporter Configuration Teess Start at Well 1 Sp Order By Row Veiw Log 0						

Sipper Settings

Steps	Details					
5 Home the cartridge changer and replace	Click System Tools > Cartridge Changer . Refer to the <i>User Guide</i> for more details.					
ourthage	🔀 Cartridge Changer					
	Configuration Parameters					
	V Auto Cartridge Switch					
	Pump1 Pressure Limit (MPa) 18					
	Pump3 Pressure Limit (MPa) 18					
	Type Sample Count					
	Cartridge 1 A or C4 982					
	Cartridge 2 A2 or C4 other size 0					
	Catridge 3 A large bed 12uL 0					
	Catitidge 4 0 0 cyano 0					
	Catridge 6 D or graphitic carbon 41					
	Cartridge 7 E or C8 20					
	Cartridge 8 For phenyl 0					
	Cartridge 9 H1 or HILIC 0					
	Cartridge 10 O or blank empty 0					
	Cartridge 11 G9210A custom1 0					
	Cartridge 12 G9210A custom2 51					
	Homed					
	Go To Load • 12 234					
5 Home the stages	 a Install an empty example plate in either the 96- or 384-well format. b Click System Tools > Sipper Configuration to open the Sipper Configuration wizard, 					
	Refer to the User Guide for details.					
	Sipper Configuration Wizard					
	Please select plate to configure					
	Unselected					
	Unselected 384 Standard Plate					
	96_DeepWellConicalBottom_P					

Getting Started

System Preparation

System Preparation

Steps	Details					
7 Check the valves	Click System Tools > Valve Tuner to open the Valve Configuration Utility dialog box. Refer to the <i>User Guide</i> for details.					
	🕸 RapidFire Valve Configuration Utility					
	Valve 1 Positions Cur772 Load E22 Set Inj770 Clear Set Find					
	Valve 2					
	Positions Home Cur. -1020 Load Set Irnj. -1020 Clear Set Find					
	Valve 3 Home					
	Cur. 108 Load 390 V Limited Homing V Set Inj. 106 Clear Set Find					

All Valves

Load

Inject

Don't Change

V1 V2

• •

• •

0 0

0

0

8 Prepare the peristaltic pump

Check that Pump 4 is active. Make sure the pump rotates clockwise.

V4

•

 \odot

Load

Inject

Update

V3 •

۲

0 0

0 0

Refresh

1.5	0	mL/min	1.25	0	mL/min	1.25	0	mL/min	
10	10	B%	0	0	B%	80	80	B%	
0	0	C%	0	0	C%	0	0	C%	
0	0	D%	0	0	D%	0	0	D%	
0.03		MPa	0.03		MPa	0.03		MPa	

Steps	Details					
9 Prepare the quaternary pumps	<image/>					
10 Flush the sipper tube	Under Sipper Settings, click Flush Now. Sipper Settings V Wash Needle Between Sips Sip Height (mm) 2 Blank Injections Between Wells 0 Flush Now Vac Pressure (kPa)					
11 Prepare the Agilent MassHunter Data Acquisition program.	 a Click the RapidFire Communicator on the desktop. b Start the Data Acquisition program. c Turn on the detector. For Q-TOF only d Calibrate the TOF. e Create or edit a method with Q-TOF tab settings that are appropriate for your analysis. Save the method 					

Run RapidFire in Plates Mode

epare plates. ad plates.	Thaw sample plates a minutes. Manually load the pla Plate Info Current Plate Name Plates Completed	and spin on tes or use th	a centrifuge ne BenchBot	at 3000 rpm for at least § plate handler.
ad plates.	Manually load the pla Plate Info Current Plate Name Plates Completed	tes or use th	ne BenchBot	plate handler.
	Input Stack		0 NA	
	Current Column Position		Unknown	
	Plate Configuration	StandardPlate	_96 🔻	
		StandardPlate	_96	
	Miscellaneous Settings			
	Plates Between Flushes	1	4	
	Missed Sip Tolerance	5	10	
	epare the MassHunter ta Acquisition program.	Plate Configuration Plate Configuration Miscellaneous Settings V Use Plate Handle Use Barcode So V Mass Spec Stan Plates Between Flushes Missed Sip Tolerance epare the MassHunter ta Acquisition program.	Plate Configuration StandardPlate StandardPlate Miscellaneous Settings V Use Plate Handler Use Barcode Scanner V Mass Spec Standby After Run Plates Between Flushes 1 Missed Sip Tolerance 5 epare the MassHunter ta Acquisition program. a For Triple Quad LC/MS, open of on the QQQ tabs for your analy	Plate Configuration StandardPlate_96 Viscellaneous Settings StandardPlate_96 Miscellaneous Settings V Use Plate Handler Use Barcode Scanner V Mass Spec Standby After Run Plates Between Flushes 1 Missed Sip Tolerance 5 To Triple Quad LC/MS, open or create a m on the QQQ tabs for your analysis.

Getting Started Run RapidFire in Plates Mode

parameters.	select the Plates op	Set parameters in the RapidFire dialog box for your analysis. Make select the Plates option. Save the RapidFire method.						
	🔀 RapidFire							
	File Batch Configuration Tools	System Tools Help						
	Plateset Run Controls	Well	Info					
		Tota Well	Total Wells Sipped Wells Sipped Since Reset					
	Plates Mode Settings	Cum	ent Well Address					
	Start at Well A +	Plate	Info					
	Sin Order @ Dr Dour	By Col Dista	erit mate Name					
	Sporter by how C	Input	t Stacks					
	Sipper Settings	Outp	ut Stacks					
	Cin Height (mm)	15 Curre	ent Cartridge Position					
	Sip Height (mm) Blank Injections Between Welle	Plate	e Configuration	•				
	Flush Now Vacuum Lev	el (kPa) Misce	ellaneous Settings					
	RapidFire Cycle Durations		Use Plate Handler					
	State Edit Time (ms	s) Time (m ^	Use Barcode Scanner					
	Aspirate 600	600	Mass Spec Standby After Run					
	Load/ 3000	3000 E	tes Retween Flushes					
	Extra 0	2000	read Sin Tolerance	00				
	Beeguil 500	500 *						
	Pump Settings							
	Pump 1	Pump 2	Pump 3	Pump 4				
	•	•	•					
	1.5 0 mL/min	1.25 0 mL/min	1.25 0 mL/min					
	0 0 B%	0 0 B%	0 0 B%					
	0 0 C%	0 0 C%	0 0 C%					
	0 0 D%	0 0 D%	0 0 D%					
	MPa	МРа	MPa					
	Disconnected		Apply Disconnect Conn	ect •				
	Disconnected							

Run RapidFire in Sequence Mode

The following diagram illustrates the terminology used to define a batch of plates in Sequences mode.



Steps		Details	Details					
1	Prepare plates.	es. Thaw sample plates and spin on a ominutes.			centrifuge at 3000 rpm for at least 5			
2	Load plates.	Manually load the pl	ates or use th	ne Benc	hBot plate handler.			
		Plate Info Current Plate Name						
		Plates Completed		0				
		Input Stack		NA				
		Output Stack		NA				
		Current Column Position		Unkno	wn			
		Plate Configuration	StandardPlate	_96	_			
			StandardPlate	_96				
		Miscellaneous Settings						
		🔽 🗹 Use Plate Hand	ller					
		Use Barcode S	canner					
		✓ ✓ Mass Spec Sta	ndby After Run					
		Plates Between Flushe	s 1	4				
		Missed Sip Tolerance	5	10				

Getting Started

Run RapidFire in Sequence Mode

Steps

Details

4 Set RapidFire run parameters. Set parameters in the RapidFire dialog box for your analysis. Make sure to select the **Plates** option. Save the RapidFire method.



5 Edit the plate map. Click **Batch Configuration Tools > Plate Map Editor**. Create or edit a sequence for a plate map, then save the sequence.



Getting Started Run RapidFire in Sequence Mode

Steps	Details						
6 Edit the batch	Click Batch Configurati For each sequence in th the RapidFire batch.	on Tools > ne batch, as	Batch Eo ssign a R	ditor . Create or edit the batch apidFire method to run. Save			
7 Set run parameters.	Select the correct plate analysis.	configurat	on. Verif	y other parameters for your			
	Plate Info						
	Plates Completed		0				
	Input Stack		NA	-			
	Output Stack		NA				
	Current Column Position		Unknown				
	Plate Configuration	StandardPlate_S	6				
	5	StandardPlate_9	6				
	Miscellaneous Settings	◀					
	Use Plate Handler						
	Use Barcode Scan	ner					
	🔽 🗸 Mass Spec Standby	After Run					
	Plates Between Flushes	4	4				
	Missed Sip Tolerance	10	10				

State #1: Aspirate

RapidFire Integrator Flow Path

State #1: Aspirate



State #2: Wash/Load

State #2: Wash/Load



State #3: Extra Wash

State #3: Extra Wash



State #4: Elute

State #4: Elute



State #5: Re-equilibrate



Flush the sipper tube

Flush the sipper tube



Physical colors of the tubing



Valve positions



In This Book

This guide gives you a quick overview of the RapidFire 365 High-throughput Mass Spectrometry System.

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