

# **User Manual**

Agilent 1260 Infinity Analytical- and Preparative-scale Fraction Collectors





# **Agilent Technologies**

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## WARNING

A WARNING notice denotes a hazard. It calls attention to an operating procedure, practice, or the like that, if not correctly performed or adhered to, could result in personal injury or death. Do not proceed beyond a WARNING notice until the indicated conditions are fully understood and met.

## In This Guide...

This manual contains technical reference information about the Agilent 1260 Infinity Fraction Collectors analytical and preparative scale. The manual describes the following:

#### **1** Installing the Fraction Collector

This chapter provides information about the installation of the Agilent 1260 Infinity Fraction Collectors G1364B and C.

#### 2 Configuration and Operation of the Fraction Collector

This chapter describes the configuration and operation of the fraction collector including guidelines to optimize the system and to avoid problems.

#### **3** Troubleshooting and Test Functions

This chapter describes the modules built-in troubleshooting and test functions.

#### 4 Repairing the Fraction Collector

This chapter contains instructions on simple repair and maintenance procedures.

#### 5 Parts and Materials

This chapter contains lists for identification of common repair and maintenance parts.

#### 6 Specifications

This chapter contains performance specifications of the fraction collectors.

#### A Safety Information

This appendix provides a safety summary.

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Agilent 1260 Infinity Fraction Collectors User Manual

# **Installing the Fraction Collector**

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1 Installing the Fraction Collector Site Requirements

## Site Requirements

A suitable site environment is important to ensure optimum performance of the Fraction Collector.

## **Power Consideration**

The Fraction Collector power supply has wide-ranging capability (see Table 1 on page 12). Consequently there is no voltage selector in the rear of the Fraction Collector. There are also no externally accessible fuses, as automatic electronic fuses are integrated in the power supply.

The thermostatted fraction collector comprises two modules, the fraction collector (G1364B or G1364C) and the thermostat (G1330B). Both modules have a separate power supply and a power plug for the line connections. The two modules are connected by a control cable and both are turned on by the Fraction Collector module. The thermostat power supply has two externally accessible fuses.

## WARNING

To disconnect the Fraction Collector from line power, unplug the power cord. The power supply still uses some power, even if the power switch on the front panel is turned off.

## WARNING

To disconnect the thermostatted Fraction Collector from line power, unplug the power cord from the Fraction Collector and the thermostat. The power supplies still use some power, even if the power switch on the front panel is turned off. Please make sure that it is always possible to access the power plug.

## WARNING

Shock hazard or damage of your instrumentation can result if the devices are connected to a line voltage higher than specified.

## **Power Cords**

Your Fraction Collector is delivered with a power cord which matches the wall socket of your particular country or region. The plug on the power cord which connects to the rear of the instrument is identical for all types of power cord.

#### WARNING

Never operate your instrumentation from a power outlet that has no ground connection. Never use a power cord other than the power cord designed for your region.

## WARNING

Never use cables other than the ones supplied by Agilent Technologies to ensure proper functionality and compliance with safety or EMC regulations.

## **Bench Space**

The Fraction Collector dimensions and weight (see Table 1 on page 12) allow the instrument to be placed on almost any laboratory bench. The instrument requires an additional 2.5 cm (1.0 inch) of space on either side, and approximately 8 cm (3.1 inches) at the rear for the circulation of air, and room for electrical connections. Ensure the Fraction Collector is installed in a horizontal position.

The thermostatted Fraction Collector dimensions and weight (see Table 2 on page 13) allow the instrument to be placed on almost any laboratory bench. The instrument requires an additional 25 cm (10 inches) of space on either side for the circulation of air, and approximately 8 cm (3.1 inches) at the rear for electrical connections. Ensure the Fraction Collector is installed in a horizontal position.

If a complete Agilent 1260 Infinity LC System is to be installed on the bench, make sure that the bench is designed to carry the weight of all the modules. For a complete system including the thermostatted Fraction Collector it is recommended to position the modules in two stacks, see "Recommended Stack Configuration - Fraction Collector in an Analytical System (Front View)" on page 18. Make sure that in this configuration there is 25 cm (10 inches) space on either side of the thermostatted Fraction Collector for the circulation of air.

**Site Requirements** 

## Environment

Your Fraction Collector will work within specifications at ambient temperatures and relative humidity as described in Table 1 and Table 2 on page 13.

## CAUTION

Do not store, ship or use your Fraction Collector under conditions where temperature fluctuations may cause condensation within the Fraction Collector. Condensation will damage the system electronics. If your Fraction Collector was shipped in cold weather, leave it in its box, and allow it to warm up slowly to room temperature to avoid condensation.

#### Table 1 Physical Specifications - Fraction Collectors (G1364B, G1364C)

Туре	Specification	Comments
Weight	13.5 kg (29.8 lbs)	
Dimensions (height × width × depth)	200 × 345 × 440 mm (8 × 13.5 × 17 inches)	
Line voltage	100-240 VAC, ±10 %	Wide-ranging capability
Line frequency	50 or 60 Hz, ±5 %	
Power consumption (apparent power)	200 VA	Maximum
Power consumption (active power)	180 W	Maximum
Ambient operating temperature	4 – 45 °C (41 – 113 °F)	
Ambient non-operating temperature	-40-70 °C (-4-158 °F)	
Humidity	< 95 %, at 25 – 40 °C (77 – 104 °F)	Non-condensing
Operating Altitude	Up to 2000 m (6500 ft)	
Non-operating altitude	Up to 4600 m (14950 ft)	For storing the Fraction Collector
Safety standards: IEC, CSA, UL	Installation Category II, Pollution Degree 2. For indoor use only.	

Туре	Specification	Comments
Weight	18.5 kg (40.7 lbs)	
Dimensions (height × width × depth)	140 × 345 × 435 mm (5.5 × 13.5 × 17 inches)	
Line voltage	100 $-$ 120 or 220 $-$ 240 VAC, $\pm$ 10 %	Wide-ranging capability
Line frequency	50 or 60 Hz, ± 5 %	
Power consumption (active power)	210 W	Maximum
Power consumption (apparent power)	260 VA	Maximum
Ambient operating temperature	4 - 40 °C (41 - 131 °F)	
Ambient non-operating temperature	-40-70 °C (-4-158 °F)	
Humidity	< 95 %, at 25 $-$ 40 $^{\circ}$ C (77 $-$ 104 $^{\circ}$ F)	Non-condensing
Operating Altitude	Up to 2000 m (6500 ft)	
Non-operating altitude	Up to 4600 m (14950 ft)	For storing the Fraction Collector
Safety standards: IEC, CSA, UL, EN	Installation Category II, Pollution Degree 2. For indoor use only.	

## Table 2 Physical Specifications - Thermostat (G1330B)

1 Installing the Fraction Collector Unpacking the Fraction Collector

## **Unpacking the Fraction Collector**

## CAUTION

If you need to ship the Fraction Collector at a later date, always use the shipping protection foam parts (see "Transporting the Fraction Collector" on page 41).

## **Damaged Packaging**

Upon receipt of your Fraction Collector, inspect the shipping containers for any signs of damage. If the containers or cushioning material are damaged, keep them until the contents have been checked for completeness and the Fraction Collector has been mechanically and electrically checked. If the shipping container or cushioning material is damaged, notify the carrier and keep the shipping material for the carrier's inspection.

## CAUTION

If there are signs of damage to the Fraction Collector, please do not attempt to install the Fraction Collector.

## **Delivery Checklist**

Ensure all parts and materials have been delivered with the Fraction Collector. For this compare the shipment content with the checklist included in each instrument box. Please report missing or damaged parts to your local Agilent Technologies sales and service office.

Three models of Agilent 1260 Infinity fraction collectors are available:

- *G1364B* Fraction Collector, *preparative-scale*, designed for flow rates up to 100 ml / min. and for the use with vials, deep well plates and test tubes up to 100 mm height.
- *G1364C* Fraction Collector, *analytical-scale*, designed for flow rates below 10 ml / min. (can be modified for higher flow rates) and for the use with vials, test tubes of up to 75 mm height, well-plates and a 40-funnel tray connecting to external locations of any size

• *G1364C Thermostatted* Fraction Collector, *analytical scale*, can be created by additionally ordering and installing a G1330B Fraction Collector Thermostat.

The terms (thermostatted) *analytical scale* Fraction Collector and the *preparative scale* Fraction Collector refer to these Agilent 1260 Infinity modules in this manual. Unless otherwise stated all information in this section is valid for all models.

 Table 3
 Delivery Checklist for the G1364B (preparative-scale) and G1364C (analytical-scale) Fraction Collector

Description	Quantity
Fraction collector module with inlet / waste tubing assembly	1
Power cord, local (Matching the sockets in your country or region)	1
Accessory kit (see below)	1
Delay Calibrant G1946-85020	1

#### Table 4 Delivery Checklist for the G1330B Thermostat Module (optional)

Description	Quantity
Thermostat Module	1
Power cord, local (Matching the sockets in your country or region)	1
Accessory kit thermostat module (see below)	1

## **Accessory Kits**

Each shipment contents an Accessory kit with the necessary tools to install the system and to have an operating system.

**Unpacking the Fraction Collector** 

Description	Quantity	Part Number
Wrench, open end, 4mm	1	8710-1534
Wrench, open end, 1/4-5/16 inch	1	8710-0510
Hex key 2.0 mm	1	8710-2476
Finger tight fittings, 1/16″ f-120 <sup>*</sup>	3	0100-1516
Waste tubing (1.2 m) <sup>†</sup>	1	5062-2463
Y-Type Fitting for waste tubing <sup>‡</sup>	1	5065-9971
CAN cable, 1 m	1	5181-1519
Air channel adapter	1	G1329-43200
Sticking clamp for corrugated waste tubing (large)**	3	5065-9976
Sticking clamp for waste tubing (small) $^{\dagger\dagger}$	3	5065-9976
Tray for 15 x 6 ml vials	1	G1313-44513
Tray for 40 x 2 ml vials	1	G1313-44512

#### Table 5 Fraction Collector Accessory Kit Contents G1364-68725

- \* Reorder gives pack of 2
- † Reorder gives 5 m
- ‡ Reorder gives pack of 10
- \*\* Reorder gives 10 clamps for corrugated waste tubing (large))
- tt Reorder gives 10 clamps for waste tubing (small)
- The Accessory kit (G1330-68705) shown in Table 6 is shipped with the (G1330B) thermostat module if the thermostat module was ordered.

#### Table 6 (Optional) Thermostat Module Accessory Kit Contents G1330-68705

Description	Quantity	Part Number
Waste tubing (1.2 m) <sup>*</sup>	1	5062-2463
Cable thermostat module to Fraction Collector	1	G1330-81600

\* Reorder gives 5 m

## **Optimizing the Stack Configuration**

If your Fraction Collector is part of a system, you can ensure optimum performance and minimum delay volume by installing the following configuration.

- Figure 1 and Figure 2 on page 19 show the configuration recommended for the Fraction Collector in an analytical scale system.
- Figure 3 on page 20 and Figure 4 on page 21 show the configuration recommended for the Fraction Collector with a preparative scale system.

**NOTE** Agilent 1260 Infinity LC Purification Systems can comprise many different LC modules. This section describes some common stack configurations and general considerations. On more complex systems, e.g. with two or more fraction collectors or with mass selective detector, other stack configurations might show advantages.

**Optimizing the Stack Configuration** 

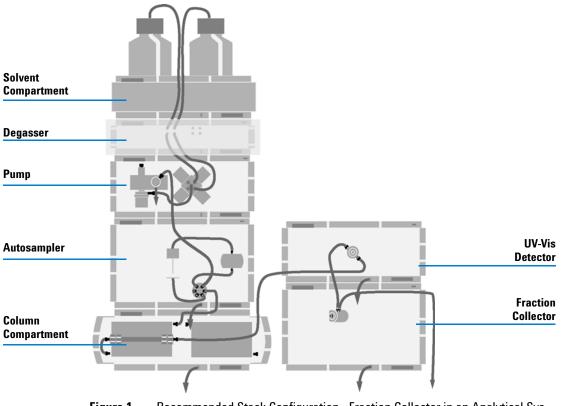


Figure 1 Recommended Stack Configuration - Fraction Collector in an Analytical System (Front View)

NOTE

For information about connecting an Agilent LC-MSD, a Non-1260 Infinity or Non-UV-Vis detector to the system, please refer to the "User's Guide for Purification / High Throughput System", PN G2262-90010 or to the "Installation Note for the Mass Based Fraction Collection Kit".

**Optimizing the Stack Configuration** 

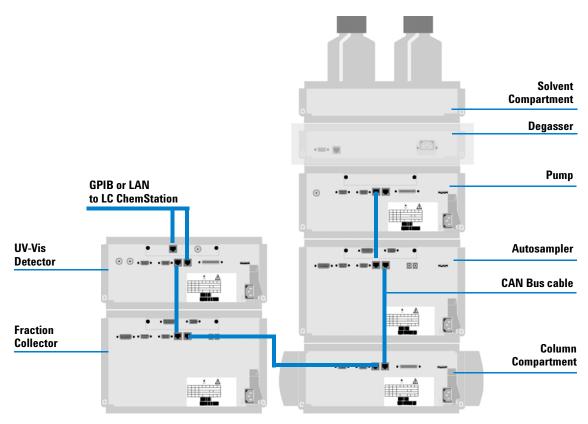


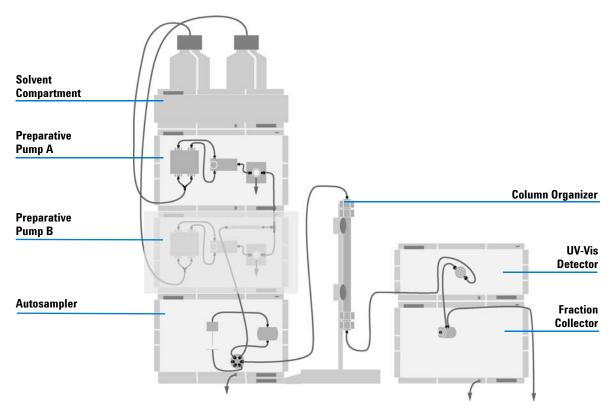
Figure 2 Recommended Stack Configuration - Fraction Collector in an Analytical System (Rear View)

**NOTE** If a G1330B ALS thermostat is part of the system it must always be installed underneath the thermostatted injector or Fraction Collector (see "Installing a Thermostatted Fraction Collector" on page 26).

NOTE

The fraction collector should never be positioned on top of a module that generates heat. This could lead to an unwanted evaporation of fractions in the fraction collector (e.g. Agilent 1260 Infinity Thermostatted Column Compartment G1316A or Agilent 1260 Infinity Diode Array Detectors.

**Optimizing the Stack Configuration** 



**Figure 3** Recommended Stack Configuration - Fraction Collector in a Preparative System (Front View)

## NOTE

For information about connecting an Agilent LC-MSD, a Non-1260 Infinity or Non-UV-Vis detector to the system, please refer to the "User's Guide for Purification / High Throughput System", PN G2262-90010 or to the "Installation Note for the Mass Based Fraction Collection Kit".

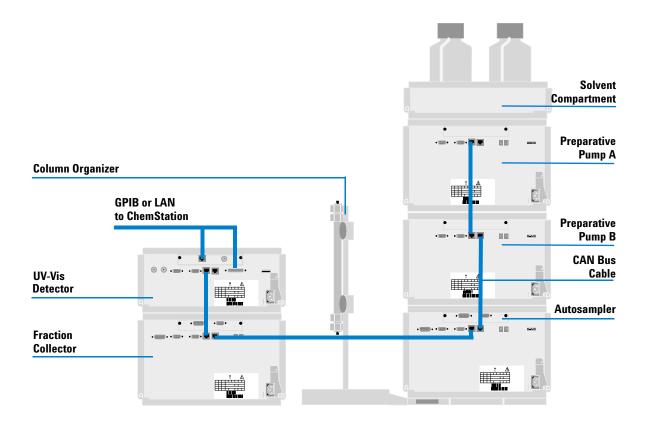


Figure 4 Recommended Stack Configuration - Fraction Collector in a Preparative System (Rear View)

NOTE

If a G1330B ALS thermostat is part of the system it must always be installed underneath the module, that is thermostatted injector or Fraction Collector (see "Installing a Thermostatted Fraction Collector" on page 26).

## NOTE

The fraction collector should never be positioned on top of a module that generates heat, which can lead to an unwanted evaporation of fractions in the fraction collector (e.g. Agilent 1260 Infinity Thermostatted Column Compartment G1316A or Agilent 1260 Infinity Diode Array Detectors.

1 Installing the Fraction Collector Installing the Fraction Collector

# **Installing the Fraction Collector**

Preparation	Locate bench space
	Provide power connections
	Unpack the Fraction Collector
Parts required	Fraction Collector
	Power cord,
	Chemstation or Agilent Instant Pilot G4208A

## WARNING

When opening capillary or tube fittings solvents may leak out. Please observe appropriate safety procedures (for example, goggles, safety gloves and protective clothing) as described in the material handling and safety data sheet supplied by the solvent vendor, especially when toxic or hazardous solvents are used.

## WARNING

To avoid personal injury, keep fingers away from the needle area during Fraction Collector operation. Do not attempt to insert or remove a vial or a plate when the needle is positioned.

- 1 Install the LAN interface board in the Fraction Collector (if required).
- 2 Remove the adhesive tape which covers the side and front doors.
- **3** Open the front door and remove the left side door.
- **4** Remove the transport protection foam.
- **5** Install the corrugated waste tube in the plastic port at the front bottom center of the instrument. Slide the waste tubing coming from the internal tray (if present) through the plastic port and the corrugated waste tube (see Figure 5 on page 23). Route the corrugated waste tubing into a waste container.

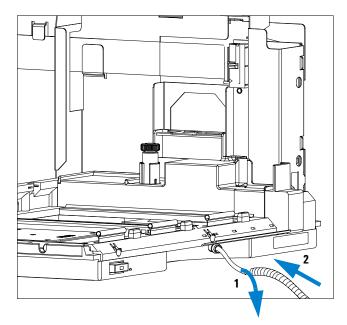


Figure 5 Installing the Corrugated Waste Tubing in the Plastic Port

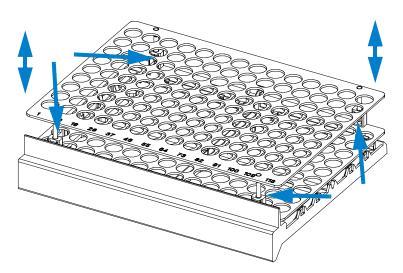
- **6** Re-install the left side door (take care of the magnet at the back). Ensure the side door is correctly installed (its presence is sensed by a hall sensor, a missing side door will result in a NOT-READY state of the instrument).
- **7** Place the Fraction Collector in the stack or on the bench in all horizontal position.
- 8 Ensure the power switch at the front of the Fraction Collector is OFF.
- **9** Connect the power cable to the power connector at the rear of the Fraction Collector.
- 10 Connect the CAN cable to the other Agilent 1260 Infinity LC modules.
- **11** If an Agilent ChemStation is the controller, it must be connect to the LAN interface (should be installed to the detector).
- **12** Connect the APG remote cable (optional) for non Agilent 1260 Infinity instruments.
- **13** Install the tray that has been delivered for your fraction collector. The test tube trays can be adjusted in height, depending on the height of the used test tubes. To adjust the height of the trays, press against the snappers at

**Installing the Fraction Collector** 

the guides in the 4 corners of a tray and move the top plate of the tray up or down (see Figure 6 on page 24).

## NOTE

The preparative-scale fraction collector (G1364B) and the analytical-scale fraction collector (G1364C) are shipped with two trays (40x2 ml half tray and 15 x 6ml half tray).



**Figure 6** Adjusting the Height of the (Full) Test Tube Trays.

- **14** Turn ON power by pushing the button at the lower left hand side of the Fraction Collector.
- **15** The exhaust fan will turn ON and remove potential solvent vapor from the inside of the instrument. After 2 minutes close the front door. Then the Fraction Collector will start the hardware initialization process. At the end of this process the status LED should be green.

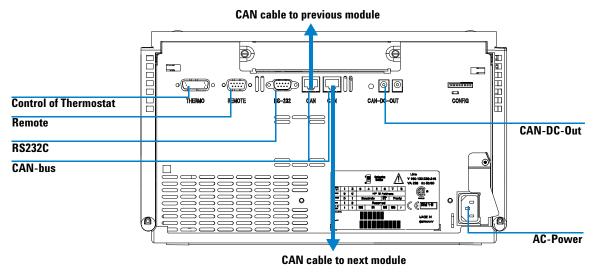


Figure 7 Cable Connections

**NOTE** The Fraction Collector is turned ON when the line power switch is pressed and the green indicator lamp is illuminated. The detector is turned OFF when the line power switch is protruding and the green light is OFF.

## WARNING

To disconnect the Fraction Collector from the line, unplug the power cord. The power supply still uses some power, even if the switch at the front panel is turned OFF.

**Installing a Thermostatted Fraction Collector** 

## Installing a Thermostatted Fraction Collector

Preparation	Locate bench space
	Provide power connections
	Unpack the Fraction Collector and the thermostat
Parts required	Fraction Collector and thermostat
	Power cords,
	ChemStation or Agilent Instant Pilot G4208A

- **1** Place the thermostat on the bench.
- **2** Remove the front cover and route the condensation drain tube to a waste container.

## WARNING

Make sure the condensation drain tube runs down into a waste container without any (upwards) bends or curves. Free and unrestricted flow of the condensation into a waste container must be guaranteed. Make sure that the condensation drain tube is always above the liquid level in the container. If the tube is located in liquid the condensed water cannot flow out of the tube and the outlet is blocked. Any further condensation will then remain in the instrument. This may damage the instruments electronics.

### CAUTION

The fraction collector thermostat requires 25 cm (10 inch) space on each for sufficient air circulation.

- **3** Install the LAN interface board in the Fraction Collector (if required).
- **4** Remove the adhesive tape which covers the side and front doors.
- 5 Open the front door and remove the left side door.
- 6 Remove the transport protection foam.
- 7 Install the corrugated waste tube in the plastic port at the front bottom center of the Fraction Collector and route down into a waste container. Slide the waste tubing coming from the internal tray (if present) through

the plastic port and the corrugated waste tube (see Figure 5 on page 23). Route the corrugated waste tubing into a waste container.

- 8 Re-install the left side door (take care of the magnet at the back). Ensure the side door is correctly installed (its presence is sensed by a hall sensor, a missing side door will result in a NOT-READY state of the instrument).
- **9** Place the Fraction Collector on top of the thermostat. Make sure that the Fraction Collector is correctly engaged in the thermostat locks.
- **10** Remove the plastic cover from the tray base, place the air channel adapter (1) into the Fraction Collector tray base. Make sure the adapter is fully pressed down. This assures that the cold airstream from the thermostat is correctly guided to the tray area of the Fraction Collector. Place the plug channel (2) on top of the air channel adapter. Both devices must be installed correctly, to assure proper operation of the instrument.

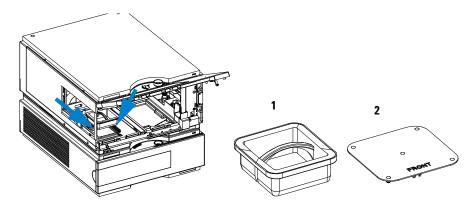


Figure 8 Installation of Thermostat and Fraction Collector

**11** The *analytical scale* Fraction Collector is delivered with a pre-installed tray compartment divider. This divider should only be used with the thermostatted Fraction Collector, if a (half or std.) tray is installed into the left and center positions of the instrument. If any tray is installed to the right side of the instrument, remove the tray compartment divider. The tray compartment divider optimizes the cooling performance of the instrument, if only the left and center position of the Fraction Collector are in use.

**Installing a Thermostatted Fraction Collector** 

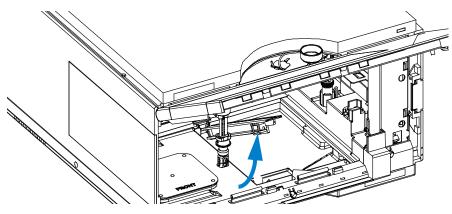


Figure 9 Removing the Tray Compartment Divider

12 Install the tray you have ordered for your fraction collector. The test tube trays can be adjusted in height, depending on the height of the used test tubes. To adjust the height of the trays, press against the snappers at the

guides in the 4 corners of a tray and move the top plate of the tray up or down (see Figure 6 on page 24).

- **13** Ensure the power switch on the front of the Fraction Collector is 0FF and the power cables are disconnected.
- 14 Connect the cable between the Fraction Collector and the thermostat, see Figure 10 on page 30.

# **WARNING** Do not disconnect or reconnect the Fraction Collector to thermostat cable when the power cords are connected to either of the two modules. This will damage the electronics of the modules.

**15** Connect the power cables to the power connectors.

- 16 Connect the CAN cable to other Agilent 1260 Infinity LC modules.
- **17** If an Agilent ChemStation is the controller, it must be connect to the LAN interface (should be installed to the detector).
- **18** Connect the APG remote cable (optional) for non Agilent 1260 Infinity instruments.
- **19** Turn ON power by pushing the button at the lower left hand side of the Fraction Collector.
- **20** The exhaust fan will turn ON and remove potential solvent vapor from the inside of the instrument. After 2 minutes close the front door. Then the Fraction Collector will start the hardware initialization process. At the end of this process the status LED should be green.
- **21** The Fraction Collector is turned ON when the line power switch is pressed and the green indicator lamp is illuminated. The detector is turned OFF when the line power switch is protruding and the green light is OFF.

## WARNING

To disconnect the Fraction Collector from the line, unplug the power cord. The power supply still uses some power, even if the power switch at the front panel is turned OFF.

## WARNING

To avoid personal injury, keep fingers away from the needle area during Fraction Collector operation. Do not attempt to insert or remove a vial or a plate when the needle is positioned.

Installing a Thermostatted Fraction Collector

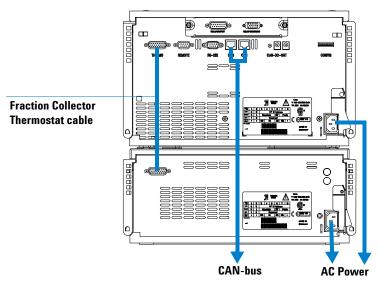


Figure 10 Connections at the Rear of the Thermostatted Fraction Collector

# **Flow Connections to the Fraction Collector**

	Preparation Parts required	Fraction Collector is installed in the LC system Parts from the accessory kit, see "Fraction Collector Accessory Kit Contents G1364-68725" on page 16
WARNING	appropriate sat clothing) as de	capillary or tube fittings, solvents may leak out. Please observe fety procedures (for example, goggles, safety gloves and protective scribed in the material handling and safety data sheet supplied by the , especially when toxic or hazardous solvents are used.
WARNING	Regularly inspect the inlet / waste tubing assembly and the valve to needle tubing and exchange them if they are worn out or show visible signs of damage.	
WARNING	inlet / waste tu or fraction loss	ow the described installation procedures to maximize the lifetime of ubing assembly the valve to needle tubing and to avoid potential spills es. Regularly inspect the tubings and exchange them if they are worn sible signs of damage.
WARNING	This instrumen higher than 200	t should only be used with solvents that have an ignition temperature D°C!

**Flow Connections to the Fraction Collector** 

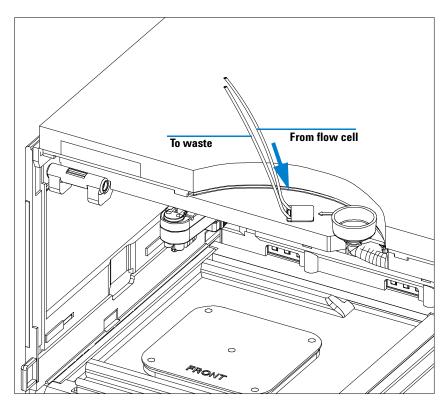


Figure 11 Flow Connections to and from the Fraction Collector.

Route the waste outlet around the instrument as shown below and into a corrugated waste tubing. Fix the waste outlet tubing and the corrugated waste tubing to the instrument with the sticking holders (part of the G1364-68725 accessory kit) as shown below. Route the corrugated waste tubing into a waste container.

Flow Connections to the Fraction Collector

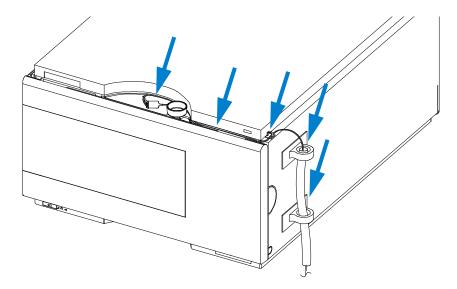


Figure 12 Fixing and Routing the Waste Tubings

1 Installing the Fraction Collector Fraction Collector Trays

# **Fraction Collector Trays**

# Installing the Fraction Collector Trays 1 Press the front door latch-holding button located at the front of the right-side cover. 2 Lift the front door. **3** Adjust the top plate of the test tube tray for the correct tube height if required. **4** Load the Fraction Collector tray with Fraction Collector well-plates, test tubes or vials as required. 5 Slide the Fraction Collector tray into the Fraction Collector so that the rear of the tray is seated firmly against the rear of the tray area. Installed trays are automatically detected and identified. NOTE **6** Press the front of the Fraction Collector tray down to secure the tray in the Fraction Collector. 7 Close the front door. If the tray pops out of position the air channel adapter is not correctly inserted. NOTE Before starting a run, the instrument has to be correctly configured in the user interface. NOTE

## Numbering of Vial, Test Tube and Well-plate Positions

#### With the 4 plates full tray

Plate in the left front position: P1 Plate in the left back position: P2 Plate in the right front position: P3 Plate in the right back position: P4 Vessel: A1; A2;... B1; B2;

#### With the 2 plates / 10 x 2ml vials or 10 funnels std. trays

Plate in the front position: P1 Plate in the back position: P2 Vessel: A1; A2;... B1; B2;... Vials / funnels: 1 - 10

#### With the 100 vials std. tray

Vial: 1 - 100

#### With the half-trays

Left-hand 40-position tray: 1 - 40 Center 40-position tray: 101-140 Right-hand 40-position tray: 201 - 240 or Left-hand 15-position tray: 1 - 15 Center 15-position tray: 101-115

Right-hand 15-position tray: 201 - 215

	With the 40, 60, 125 or 215 position test tube full trays
	Numbering starts in front left corner in columns to the back and then to the right.
WARNING	If you are using flammable solvents, remove the plates from the Fraction Collector after turning it OFF. You avoid the risk of building explosive gas mixtures in the instrument.
WARNING	If you are using flammable solvents, cover the plates. You avoid the risk of building explosive gas mixtures in the instrument.
WARNING	Closing mats with adhesive can give some contamination in the system. The adhesive is soluble in most of the solvents used in HPLC.
WARNING	In general do not use closing mats with adhesive. The Fraction Collector has no prepunch needle, therefore the adhesive will clog the needle after several injections.

1

**Installing the Fraction Collector** Fraction Collector Trays

## **Configure Well-plate Types**

If the plate you are using is not found on the "List of Recommended Plates and Closing Mats" on page 132 you may configure a custom plate. Measure the exact dimensions of the plate as marked below and enter the values in the plate configuration table of the ChemStation or the Agilent Instant Pilot G4208A.

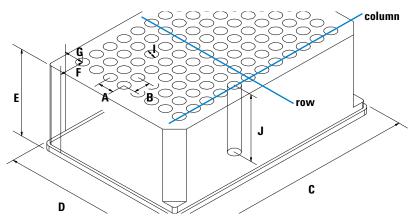


Figure 13 Well-plate Dimensions (straight)

### **1** Installing the Fraction Collector

**Configure Well-plate Types** 

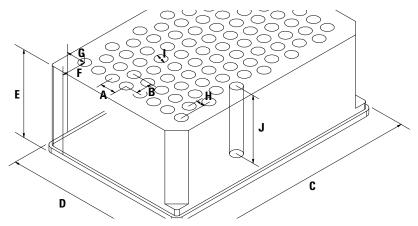


Figure 14 Well-plate Dimensions (staggered)

Location	Description	Definition	Limits
	Rows	Number of rows on the plate	up to 16
	Columns	Number of columns on the plate	up to 24
	Volume	Volume (in µl) of a sample vessel	
A	Row distance	Distance (in mm) between the center of two rows	
В	Column distance	Distance (in mm) between the center of two columns	
С	Plate length	X size (in mm) at the bottom of the plate	127.75+/- 0.25 mm (SBS Standard)
D	Plate width	Y size (in mm) at the bottom of the plate	85.50+/-0.25 mm (SBS Standard)
E	Plate height	Size (in mm) from the bottom to the top of the plate. If you use well plates with inlets, caps and septa, you have to specify the distance from the bottom to the top of the caps.	up to 47 mm
F	Column offset	Distance (in mm) from the left edge (bottom) to the center of the first hole (A1)	
G	Row offset	Distance (in mm) from the back edge (bottom) to the center of the first hole (A1)	
Η	Column shift	Offset (in mm) to Y when the rows are not straight but staggered	
J	Well depth	Distance (in mm) from the top of the plate to the bottom of the well. If you use well plates with inlets, caps and septa, you have to specify the distance from the top of the septa to the bottom of the inlets.	up to 45 mm

### Table 7 Configuring Well-plate Types

#### **1** Installing the Fraction Collector

**Configure Well-plate Types** 

Location	Description	Definition	Limits
	Well X size	Size of the well in x direction (Plate length) If you use well plates with inlets, caps and septa, you have to specify the x size of the septa.	min. 3.7 mm min. 3.0 mm with position accuracy alignment (micro scale)
	Well Y size	Size of the well in y direction (Plate width). If you use well plates with inlets, caps and septa, you have to specify the y size of the septa.	down to 3.7 mm min. 3.0 mm with position accuracy alignment (micro scale)
	Bottom size	For round wells, the relative of the top and bottom of the well	1.0: cylindrical well 0.0: conical well
	Square	Click in the field to specify whether the well is rectangular or round	Yes: rectangular No: round /oval
	ls well plate	Click in this field to specify if this is a well plate or not. Relevant for continuous flow operation.	Yes: well plate or MALDI Target No: Vial Tray or Eppendorf tray

#### Table 7 Configuring Well-plate Types (continued)

### NOTE

The distances need to be measured with high precision. It is recommended to use a caliper.

If possible contact the vendor of non-predefined well plates to obtain these dimensions.

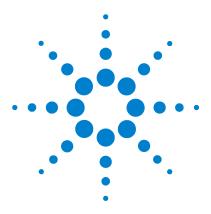
## **Transporting the Fraction Collector**

When moving the Fraction Collector inside the laboratory, no special precautions are needed. However, if the Fraction Collector needs to be shipped to another location via carrier, ensure:

- ✓ The transport assembly is in the park position. Use the ChemStation or the Agilent Instant Pilot G4208A for this command.
- To move the arm to the park position:
- Switch to the Diagnosis view of the ChemStation and select Fraction Collector
   > Maintenance Positions. from the Maintenance menu
- 2 In the upcoming dialog box click Park Arm.
- ✓ The vial tray and the sample transport mechanism is secured with the transport protection foam.

### **1** Installing the Fraction Collector

**Transporting the Fraction Collector** 



Agilent 1260 Infinity Fraction Collectors User Manual

## 2 Configuration and Operation of the Fraction Collector

Configuration of the Fraction Collector 44 Delay Volumes and Delay Calibration 50 Setting up a Fraction Collector Method 65 Starting Your Run with Fraction Collection 72 Viewing Your Results 76 Special Applications 78 Limitations and How to Avoid Problems 83 Check-out Procedures 84 Application Notes 89



Agilent Technologies

## **Configuration of the Fraction Collector**

### **Configuration of the Fraction Collector in the ChemStation**

### CAUTION

Before using the preparative-scale fraction collector G1364B the delay calibration adapter G1364-87301 has to be removed from the needle carrier assembly.

In order to setup or change the configuration parameters of your fraction collector select **More Fraction Collector > Configuration** from the Instrument menu or right-click on the fraction collector icon in the graphical user interface. In the resulting **Fraction Collector Configuration** dialog box (Figure 15 on page 45) the configuration of the **Trays**, the **Fraction Delay Volumes**, the **Collection Order**, the **Needle Movement** and the **Well-Plates** can be specified.

#### Trays

In the online mode of the Agilent ChemStation the type of the installed tray is recognized automatically. In the off-line mode an appropriate tray type can be chosen from a dropdown list. For a selection of supported trays see "Supported Trays for Fraction Collectors" on page 126.

**Tube volume [ml]** Volume of the test tube should be specified 10% less than the maximum fill volume to avoid overfill.

**Tube height [mm]** The exact test tube height has to be specified. For preparative scale fraction collector the minimum height is 48 mm and the maximum height is 100 mm. For the analytical scale fraction collector with 50 mm needle (standard) the maximum height is 48 mm. By changing to the short 20 mm needle the maximum tube height can be in creased to 75 mm. For the part numbers of the different needle assemblies see "Needle Assemblies" on page 136.

#### NOTE

In the online ChemStation the installed tray is recognized and the default settings for tube volume and tube height are loaded and displayed.

**Configuration of the Fraction Collector** 

Tray A: 40 Tubes				
Тгау В:		Tube Volume (r	nl]: 45.00	
Тгау С:		Tube Height (m	m]: 98	
raction Delay Volumes	Collection Order			
Detector         Volume(μl)           1         DAD1         500	O Row by row		<ul> <li>Shortest path by column</li> </ul>	$\mathbb{M}$
	Column by column		○ Shortest path by row	
ollection Mode	Need	le Movement		
Discrete fractions	0	into location	Depth: 5.	0 mm
O Continuous flow	۲	above location	Distance: 2.	0 mm
	0	contact contro	Distance: 0.	0 mm
/ell-Plates				
Plate Type:		atas Reserv 196	ved Locations	
Installed Plates		Row(s	):	
🗖 Plate 1 (front left) 🗖 Pla	te 3 (front right)	Colum	n(s):	
Plate 2 (back left)	ate 4 (back right)	Single Locati		

Figure 15 Fraction Collector Configuration dialog

**Use for Sample Recovery** This checkbox only appears, if multiple fraction collectors are configured (see Figure 34 on page 79). Then the last fraction collector can be selected for sample recovery. If four fraction collectors are configured, the fourth one will automatically used for sample recovery. To change the order of your fraction collectors select **Configure 1260 Access** from the **Instrument** menu. In the **Configuration** dialog box use the **Up** or **Down** button to change the configuration order and restart your ChemStation.

#### 2 Configuration and Operation of the Fraction Collector Configuration of the Fraction Collector

#### **Fraction Delay Volumes**

The Fraction Delay Volume specifies the volume between the detector cell and fraction collector diverter valve. This volume has to be determined during the delay calibration. See "Delay Volumes and Delay Calibration" on page 50 for a detailed description.

#### **Collection Order**

The Collection Order describes the way of capillary movement during fraction collection. Four different settings are possible:

- row-by-row
- column-by-column
- shortest way by row
- · shortest way by column

Shortest way by row/column is recommend, if **Continuous flow** is selected as Collection mode.

#### **Collection Mode**

The Collection Mode defines the triggering of the diverter valve. For **Discrete fractions** the diverter valve switches to waste while the needle moves to the next test tube, vial or well. For **Continuous flow** the diverter valve is not switching during needle movements, except for long movements in the row-by-row or column-by-column mode. In addition the needle cannot move **Into location** during continuous flow operation. **Continuous flow** is only available for well plates.

#### **Needle Movement**

**Into location** In the Into Location mode the needle tip moves into the well to the specified **Depth** (in mm). This option is only available for the analytical scale fraction collector in the **Discrete fractions** mode.

**Above location** In the Above Location mode the needle tip stays at the specified **Distance** (in mm) above the well during fraction collection.

**Contact Control** In this mode the needle tip moves down to the well bottom until is reaches the specified **Distance** (in mm) between needle tip and the vial/well bottom. This ensures that the forming droplet is in contact to the

vial/well bottom. During the continuing filling process the needle tip moves upwards while staying in contact with the surface of the collected liquid. This option is recommend for low flow rates and small fraction volumes to avoid air bubbles and accomplish an accurate fraction volume. It is only available for the analytical scale fraction collector.

#### **Well-Plates**

In the **Well-Plates** section the type of well plates used in a well plate tray can be configured. The well plate type can be chosen from the Plate Type dropdown list. More detailed information about all pre-configured well plates can be found in the Instrument menu. Only one type of well plate can be used on the well plate tray.

The checkbox **Treat as 4 \times 96** is available only for 384 well plates and allows to split the 384 well plate virtually into 4 separate 96-well plates. This only changes the collection order. The numbering of the wells remains as indicated on the plate. The four virtual 96 well plates start at locations A1, A13, I1 and I13, respectively.

The filling order of each quarter is as specified in the Collection Order section. When the 384 well plate is split into four equal quarters the order of the four plates is the same as displayed in the Installed Plates section.

In the **Reserved Locations**, you have the possibility to specify locations that will not be used for Fraction Collection (see Table 8).

Syntax	Description
А	Row A can't be used
A,B	Rows A and B can't be used
A-D	A, B, C and D can't be used
A-D,F	Rows A, B, C, D and F can't be used
1	Column 1 can't be used
1,2	Columns 1, and2 can't be used
1-4	Columns 1, 2, 3 and 4 can't be used
	A A,B A-D A-D,F 1 1,2

**Table 8** Syntax for the definition of Reserved Locations

**Configuration of the Fraction Collector** 

Location	Syntax	Description
	1-4,12	Columns 1, 2, 3, 4 and 12 can't be used
Single locations	G12,H12	Locations G12 and H12 can't be used

 Table 8
 Syntax for the definition of Reserved Locations (continued)

### **Configuring Multiple Fraction Collectors**

To increase the capacity of the systems up to three fraction collectors can be configured by using the Agilent 1260 Infinity 12-position13-port valve G1160A. In addition a fourth fraction collector can be configured for recovery collection.

- The **Configuration** has to be edited for all fraction collectors. The last fraction collector in the configuration can be selected for sample recovery. To configure the recovery fraction collector please read "Sample Recovery" on page 79.
- With multiple fraction collectors a fraction start location has to be specified to define the starting position. For further details see "Assignment of Start Location for Fraction Collection" on page 72. Never used the valve settings to define a start location on systems with multiple fraction collectors.
- We recommend to use inlet tubing of the same length for all fraction collectors. Otherwise a **Delay Calibration** has to be performed for each of those fraction collectors. For the recovery fraction collector the delay volume parameter will be ignored.

### NOTE

The **Delay Calibration** can only be performed for fraction collector 1 of your configured system.

The calculated delay volume is used for all fraction collectors; therefore, the volume (tubing) between the selection valve and each installed fraction collector must be identical.

Otherwise fraction collector 2 and fraction collector 3 have to be temporarily configured as fraction collector 1 during the delay calibration.

- The fraction collector inlet tubings have to be connected to port 1-3 of the selection valve in the same order as configured in the ChemStation. The recovery fraction collector is always connected to the waste tubing of the main fraction collectors. The waste tubing from multiple fraction collectors and the inlet tubing to the recovery collector have to be connected through a T-Piece.
- Fraction Collector 4 will always be used as a recovery fraction collector.

## **Delay Volumes and Delay Calibration**

### **Delay Times and Volumes**

Figure 16 shows a schematic drawing of the flow path between the detector and the fraction collector with the two delay volumes  $V_{D1}$  and  $V_{D2}$ . For peak-based fraction collection the system delay times  $t_{D1}$  and  $t_{D2}$  can be calculated by dividing the delay volumes by the flow rate  $\dot{\nu}$ .

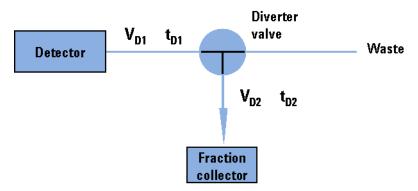


Figure 16 Delay Volumes and delay times

The delay volume  $V_{\rm D2}$  is a system parameter, it depends on the installed fraction collector tubing and the fraction collector needle. It is approximately 23  $\mu l$  for the fraction collector AS and approximately 120  $\mu l$  for the fraction collector PS. Delay volume  $V_{\rm D1}$ , which is specified in the Fraction Collector Configuration window, is determined using the Delay Volume Calibration feature of the ChemStation software.

When a peak is detected during a purification run (Figure 17) the diverter valve is triggered using the following delay time calculations:

Start of fraction collection:  $t = t_0 + t_{D1}$ End of fraction collection:  $t = t_E + t_{D1} + t_{D2}$ 

**Delay Volumes and Delay Calibration** 

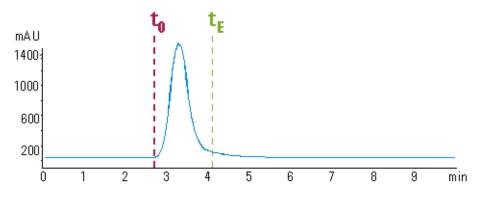


Figure 17 Chromatogram with peak start t0 and peak end tE

### **Detector Signal Delay**

Every Agilent 1260 Infinity detector that is used for triggering fractions has an internal signal delay caused by filtering the raw data. The signal delay depends on the *Peakwidth* setting of the detector and is accounted for when the diverter valve is triggered. Table 9 and Table 11 list the internal signal delay times for different Peakwidth settings.

Peakwidth (min)	Response time (sec)	Signal delay (sec)
<0.01	0.1	0.05
>0.01	0.2	0.15
>0.03	0.5	0.5
>0.05	1.0	1.25
>0.10	2.0	2.75
>0.20	4.0	5.9
>0.40	8.0	11.9
>0.85	16.0	23.9

Table 9 Signal delay times for the Agilent 1260 Infinity DAD/MWD (G1315D/G1365D)

**Delay Volumes and Delay Calibration** 

Peakwidth (min)	Response time (sec)	Signal delay (sec)
<0.0025	0.02	0.0375
>0.0025	0.05	0.0625
>0.005	0.1	0.144
>0.01	0.2	0.294
>0.03	0.5	0.619
>0.05	1.0	1.27
>0.1	2.0	2.57
>0.2	4.0	5.17
>0.4	8.0	10.4
>0.85	16.0	20.8

 Table 10
 Signal Delay Times for the Agilent 1260 Infinity DAD/MWD (1315C/G1365C)

 Table 11
 Signal Delay Times for the Agilent 1260 Infinity VWD G1314B

Peakwidth (min)	Response time (sec)	Signal delay (sec)
<0.005	<0.12	0.07
>0.005	0.12	0.14
>0.01	0.25	0.29
>0.025	0.5	0.58
0.05	1	1.3
0.1	2	2.8
0.2	4	6.0
0.4	8	12.3

Peakwidth (min)	Response time (sec)	Signal delay (sec)
<0.00125	<0.02	0.0182
>0.00125	0.02	0.0364
>0.0025	0.05	0.0728
>0.005	0.12	0.146
>0.01	0.25	0.328
>0.025	0.5	0.710
>0.05	1	1.49
>0.1	2	3.08
>0.2	4	6.26
>0.4	8	12.6

 Table 12
 Signal Delay Times for the Agilent 1260 Infinity VWD G1314C

 Table 13
 Signal Delay Times for the Agilent 1260 Infinity FLD G1321A

Peakwidth (min)	Response time (sec)	Signal delay (sec)
<0.005	<0.12	0.018
>0.005	0.12	0.073
>0.01	0.25	0.18
>0.03	0.5	0.44
>0.05	1	0.96
>0.1	2	2.0
>0.2	4	4.2
>0.4	8	8.6

**Delay Volumes and Delay Calibration** 

### CAUTION

If the internal signal delay is longer than the delay time t<sub>D1</sub> some of the peak will be lost. The maximum allowed signal delay time can be calculated using the following equation:

Signal delay time<sub>(max)</sub> =  $\frac{V_{D1}}{\dot{v}}$   $\dot{v}$  = Flow rate

After calculating the maximum signal delay time a **Peakwidth** setting can be selected that gives a signal delay time, which is shorter than the calculated maximum signal delay time. This **Peakwidth** setting should then be used for the LC purification run.

### NOTE

We recommend to set the **Peakwidth** always to > 0.01 for the DAD and MWD or to > 0.005for the VWD.

If the **Peakwidth** setting cannot be reduced and the signal delay time is longer than  $t_{D1}$  it is also possible to increase V<sub>D1</sub> by adding additional tubing. However this higher delay volume will increase the peak dispersion between detector and fraction collector.

The stop-time of the run in the ChemStation must be set to at least:

Total duration of time table (time of last entry OFF) + fraction collector delay time  $(V_{D1}/\dot{v}) + 0.1 min$  for time-based fraction collection.

End of last peak (t<sub>E</sub>) + fraction collector delay time  $(V_{D1}/\dot{v}) + 0.1 min$  for peak-based fraction collection

### Performing a Delay Calibration with an UV Detector

- **1** Place a vial containing the Delay Sensor Calibrant (G1946-85020) in position 1 of the Autosampler.
- **2** Remove the installed column and connect the capillaries with a zero-dead-volume connector.
- **3** Install the 40 x 2ml vial tray (analytical scale) or the 15 x 6ml vial tray (preparative scale) in the fraction collector.
- 4 Connect a bottle of water to Channel A.
- 5 Switch to Diagnosis View (if necessary).
- **6** Open the AFC Delay Volume Calibration status window (see Figure 18) from the Fraction Collector sub-menu of the Maintenance menu:

#### Maintenance > Fraction Collector > Delay Volume Calibration

AFC Delay Volume Calibr	ation - Status Window		×
Info			
Turn on all pumps			
	Continue	Cancel	Help
	Continue		neth

Figure 18 Delay Volume Calibration status window

7 The Info field gives the instruction to turn ON the pumps (see Figure 18). Click **Continue**.

**Delay Volumes and Delay Calibration** 

AFC Delay Volume Calibration - Status Window		×
Calibration method: DGCALAS.M		
Info I		
Change method Continue	Cancel	Help

Figure 19 AFC Delay Volume Calibration Window

When you have turned ON the pumps, click  $\ensuremath{\textit{Continue}}$  again to start the delay volume calibration.

The method to be used for the delay volume calibration is displayed (DGCALAS.M is the default calibration method for the analytical scale fraction collector and DGCALPS.M is the default method for the preparative fraction collector); if you want to change to a different method, click the Change Method button and select a new method. See also "Making Your Own Calibration Method" on page 64

The Info field shows the current status of the delay volume calibration, and gives instructions and warnings.

**8** Install the delay calibration adapter G1364-87301 by attaching it to the needle carrier assembly (G1364B preparative-scale only).

The selected method is loaded, and the delay volume calibration sample is injected.

- **9** After the delay calibration has been finished, you are prompted to remove the delay calibration adapter (G1364B preparative-scale only).
- **10** When the calibration run has finished, click **OK**. The Calibration Results panel (see Figure 20) is displayed.

**Delay Volumes and Delay Calibration** 

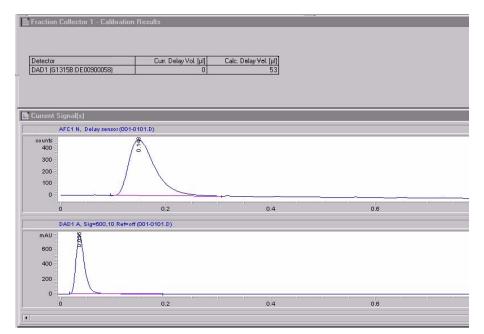


Figure 20 Delay Calibration Results Panel

The Calibration Results panel contains a results table and the acquired chromatograms from the detector(s) and the fraction collector delay sensor. The results table contains

- the name and serial number of the detector,
- the current delay volume (stored in the fraction collector configuration),
- the calculated delay volume from the calibration.

To print the Calibration Results panel click Print.

**11** If you want to store the calculated delay volume in the fraction collector configuration, click **Calibrate**. A confirmation dialog box is displayed when the delay volume has been stored; to close the confirmation click **OK**.

If you do not want to store the calculated delay volume in the fraction collector configuration, click the Close button. A confirmation dialog box is displayed; click **OK** to close, the **Confirmation** or **Cancel** to return to the Calibration Results panel.

**Delay Volumes and Delay Calibration** 

# **NOTE** The **Delay Calibration** can only be performed for fraction collector 1 of your configured system.

The calculated delay volume is used for all fraction collectors; therefore, the volume (tubing) between the selection valve and each installed fraction collector must be identical.

Otherwise fraction collector 2 and fraction collector 3 have to be temporarily configured as fraction collector 1 during the delay calibration.

### CAUTION

Before using the preparative-scale fraction collector G1364B the delay calibration adapter G1364-87301 has to be removed from the needle carrier assembly.

### Performing a Delay Calibration with an MSD

- **1** Place a vial with the Delay Sensor calibrant (G1946-85020) in position 1 of the Autosampler.
- **2** Remove the installed column and connect the capillaries with a zero-dead-volume connector or a mixer (Part No. 79835-87330).
- **3** Install the 40 x 2ml vial tray (analytical scale) or the 15 x 6ml vial tray (preparative scale) in the fraction collector.
- 4 Connect a bottle of water to Channel A of the main pump.
- **5** Connect a bottle of water with 0.1% acetic acid or ammonium formate to the make-up pump.
- **6** Load the method DGCALAS.M (analytical scale) or DGCALPS.M (preparative scale) and adjust the flow in the main pump to the flow you will be using for the analysis.

**CAUTION** Unlike for UV detectors, the delay calibration for the MSD needs to be performed whenever the flow rate is changed.

- **7** Set the flow of the make- up pump to the flow rate you will be using for your analysis.
- 8 Set the active splitter to the split ratio as used during operation.
- 9 Save the method to a new name.
- **10** Go to Diagnostics menu; select Delay volume Calibration from the Fraction Collector Sub-menu within the Maintenance menu.
- **11** Press **Change method...** to select the method you created. The new method name appears on the info line.
- 12 Start the active splitter.
- 13 Press Continue to execute the Delay Volume Calibration
- **14** Install the Delay Calibration Adapter G1364-87301 by attaching it to the needle carrier assembly (G1364B preparative-scale only).

**Delay Volumes and Delay Calibration** 

The selected method is loaded and the Delay Calibration Sample Injected.

**15** After the delay calibration has been finished, you are prompted to remove the delay calibration adapter (G1364B preparative-scale only).

16 When the Calibration Run is finished, press OK.

17 Examine the value for the MSD Calculated Delay time.

The delay time should be at least 5 seconds but may, in fact, be negative (see Figure 21).

Print	Close	Calibrate
Detector	Curr. Delay Vol. [µl]	Calc. Delay Vol. [µl
DAD1 (G1315A:US64400562)	883	349.14
Detector	Curr. Delay Time [sec]	Calc. Delay Time (sec
MSD	2	-0.294

Figure 21 Delay Volume Calibration results

**18** If the measured delay time is greater than 5 seconds, you can skip to step 23, otherwise, continue with step 19.

### CAUTION

If the measured delay time is significantly greater than 5 seconds, you should continue with the calibration at step 16 to avoid losing fractions at the end of the run.

- **19** Determine how much additional time is needed to create a 5- second delay. For example, if the time displayed is -1 sec, a six-second delay is needed.
- **20** Using the main pump flow rate, determine the desired additional delay volume. For example, at 3 ml/min and a six-second delay, an additional  $3.0 \ge 6/60$  or  $0.3 \ge$  ml is required.
- **21** Cut off a sufficient piece of the supplied ETFE tubing (G1968-60500) to provide the needed volume. The tubing's volume is 8.1  $\mu$ /cm. In this case, 37 cm would be required.
- **22** Add the tubing between the splitter and the fraction collector or, if multiple fraction collectors are installed, between the splitter and the G1160A valve.
- **23** Perform the calibration again. Verify the time is sufficient (>5 seconds, see Figure 22).

**Delay Volumes and Delay Calibration** 

Print	Close	Calibrate
Detector	Curr. Delay Vol. [µl]	Calc. Delay Vol. [µl
DAD1 (G1315A:US64400562)	883	937.14
Detector	Curr. Delay Time [sec]	Calc. Delay Time (sec.
MSD	2	11.991

Figure 22 Delay time greater than 5 seconds

- **24** Press **Calibrate** if you want to store the new calculated delay volume for the UV detector in the Fraction Collector Configuration (see also "Configuration of the Fraction Collector in the ChemStation" on page 44).
- 25 Press Print to get a hard copy of the report.
- 26 Stop the splitter.
- **27** Return to the **Method and Run Control View** and load the method you will be using to run your samples. Select Fraction Collection from the **More MSD...** sub-menu under the **Instrument** menu

Press **Parameters** and enter the final calculated delay time that appears in the report in the Collector Delay field, first converting it to minutes. In this case you would enter 11.991/60 = 0.20 minutes (see Figure 23).

**Delay Volumes and Delay Calibration** 

action Collection Parameters		
Model Type FC Hardware: Trigger Type: Pos	Other sitive Step	Co <u>n</u> figuration
Peak Timing		
M <u>i</u> n Peak Width (min): M <u>a</u> x Peak Width (min):		Time Limit (min): 15.00
MSD m/z Detection		
Peak Slope (counts/sec):	500	<u>C</u> ollector Delay (min): 0.20
Threshold (counts):	2000	Mass <u>W</u> indow (+/-): 1.0
A	nalog Output	Range (kilo counts/volt): 2000
Other Detector		
P <u>e</u> ak Slope (mv/sec):	10	Collector <u>D</u> elay (min): 0.01
Thresh <u>o</u> ld (%):	5.0	Eull Scale (volts): 0.1
ОК	Can	cel Help

Figure 23 Fraction Collection Parameters dialog box with delay volume entry

28 Save the method.

### NOTE

If you anticipate running samples at a variety of flow rates, it is best to calibrate at the highest likely flow rate first and install the appropriate length tubing. This way, there will be sufficient delay for the lower flow rates and new lengths will not need to be cut.

**Delay Volumes and Delay Calibration** 

### CAUTION

Before using the preparative-scale fraction collector G1364B the delay calibration adapter G1364-87301 has to be removed from the needle carrier assembly.

### **Making Your Own Calibration Method**

You make your own calibration method by editing one of the default calibration methods:

- **1** Load the default calibration method: DGCALAS.M in the case of an analytical system and DGCALPS.M in the case of a preparative system.
- **2** Change the method parameters (flow, runtime, solvent composition, injection volume or detector parameters) to suit your adapted calibration procedure.
- 3 Save the method with a new name in the method folder for your instrument.
- **4** Follow the appropriate procedure as described in the previous sections for running the method.

## **Setting up a Fraction Collector Method**

In order to setup or change the method parameters of your fraction collector select **Setup Fraction Collector** from the **Instrument** menu or right-click on the fraction collector icon in the graphical user interface. This will open the **Setup Fraction Collector** dialog box as displayed in Figure 24. In the **Setup Fraction Collector** dialog box general method settings are specified.

p F	action Co	llector : LC113					
acti	ion Trigge Off (	r Mode 🗋 Use Timetable	Peak-based		Max. Peal	k Duration 0.5 mi	n
1	○ all s	Working Mode Threshold/Slope s collected when a selected peak dete east one selected p	5.00 peak is detected by ctors peak detector	10.00 r: 🗖 Use	10.000		mAU
	Time	e Trigger Mod		tion [min] # of Fract			Append Cut Copy Paste
_	optime: osttime:	as Pump → min 2.50 → min Off → min		raction Collection Ne the start of collection tween fraction collect		Auxiliary Max. fill volume pe as configured Store temperatu Store delay sen	ml re signal sor signal
						T Store Orb signa	

Figure 24 Set up Fraction Collector dialog box

#### 2 Configuration and Operation of the Fraction Collector Setting up a Fraction Collector Method

## **Fraction Trigger Mode**

**Use Timetable** Enables the **Timetable** 

**Peak-based** If Peak-based is selected, the collection of a fraction is triggered by the signal of the detector, e.g. the Agilent 1260 Infinity Diode Array Detector or variable wavelength detector. The detailed trigger conditions are specified in the **Peak Detectors** table. In the peak-based trigger mode all entries in the Timetable are ignored.

**Max. Peak Duration** Defines a maximum collection time in case that the signal does not reach the condition to cut the fraction as exhibited in Figure 25. The could be caused by tailing peaks or if the baseline is drifting during gradient runs. The default value is set to 0.5 minutes. If broad peaks are expected, this value should be increased without exceeding the run time.

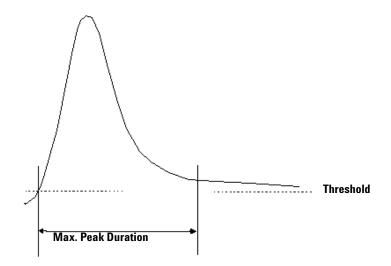


Figure 25 Maximum Fraction Duration

#### **Peak Detectors**

In the **Peak Detectors** section a list of all Peak Detectors that are connected to the system is displayed. Agilent 1260 Infinity Diode Array Detectors, multi-wavelength detectors, variable wavelength detectors and fluorescence

detectors are recognized automatically. Other Detectors, e.g. Agilent 6000 Series mass-selective detectors or HP1050 Detectors, are connected through the Universal Interface Box (UIB).

The Peak detector table contains seven columns:

Working Mode For each peak detector Threshold only, Threshold/Slope or Slope only are possible.

In the **Threshold only** mode the settings for Up Slope, Down Slope and Upper Threshold in the subsequent columns are ignored. Fraction collection is triggered whenever the detector signal exceeds the specified threshold value. When the signal drops below the threshold value fraction collection is stopped.

In the **Slope only** mode fraction collection is triggered on the slope of the detector signal. Adequate values for Up Slope and Down Slope can be specified in the corresponding fields.

In the **Threshold/Slope** mode fraction collection is triggered on the corresponding values for threshold and slope. The fraction collection is started if the detector signal exceeds both the threshold and the Up Slope value. The fraction collection is stopped if the detector signal drops either below the threshold or the Down Slope value.

To specify the trigger values **Up Slope**, **Down Slope**, **Threshold** and **Upper Threshold** we recommend to use the **Fraction Preview** tool as described in "Fraction Preview" on page 70.

**Upper Threshold** At high absorbance values the light intensity on the detector is extremely low and consequently detector noise will be superimposed on the detector signal. In this case the detector noise might trigger fraction collection. To avoid false fraction collection triggering, we recommend setting an **Upper Threshold** well below the limit where this false triggering effect might occur. As soon as the detector signal exceeds the **Upper Threshold**, settings for Up Slope or Down Slope will be ignored until the signal drops again below the Upper Threshold.

When using more than one peak detector fraction collection can be triggered either when **all selected peak detectors** detect a peak or when **at least one selected peak detector** detects a peak basing on the settings in the Peak Detectors table above. If an MSD is used for mass-based fraction collection, **Use MSD for mass-based Fraction Collection** must be checked.

#### Timetable

The **Timetable** can be used to program changes in the Fraction Trigger Mode during the analysis by entering a **Time** and specifying the trigger settings.

**Trigger Mode** Off, Peak Based and Time Based can be selected. If the Off is selected, no fractions are collected. The last entry in the timetable has to be the command Off.

Whenever the **Peak Based** mode is specified fractions will be collected based on the peak detection parameters given in the **Peak Detector** table. Additionally a **Maximum Peak Duration** in minutes has to be specified. This parameter is mandatory if you use Peak Controlled fraction collection, but is disabled for Time Based fraction collection.

When the Time Based mode is chosen two different options are available:

- The **# of Fractions** can be edited to collect a fixed number of equal fractions in a give time interval. This time interval is defined by the time value in the current and following timetable line.
- **Timeslices** [min] can be edited to collect fractions with a defined collection time. With this option the collection time of the last fraction can be shorter. This depends on the overall runtime.

For editing the Timetable the functions **Insert**, **Append**, **Cut**, **Copy** and **Paste** are offered.

To access the additional sections in the  ${\it Setup\,Fraction\,Collector}$  dialog box click More.

#### Time

In the time section of the dialog box the **Stoptime** and the **Posttime** for the fraction collector can be specified. By default the Stoptime is set to as pump and the posttime is switched OFF.

#### **Rinse Fraction Collection Needle (Analytical Scale only)**

If Discrete Fractions is selected as Collection Mode (see also"Collection Mode" on page 46), you can setup a needle rinse step before the fraction collection and/or between fractions. Then the needle will move to the

funnel on the internal tray and the diverter valve will switch to flush the needle in order to avoid carry over from the previous fraction. The instrument will determine, if it is possible to rinse the needle before the next fraction is expected.

If you have recovery positions in your fraction collector or if you are using one fraction collector for sample recovery in a multiple fraction collector configuration, the function **rinse between fractions** is ignored.

#### Auxiliary

In the Auxiliary section the **Maximum fill volume** per location can be specified. If **as configured** is selected, the pre-configured volume (see **Instrument > Pre-configured Wellplate Types**) is used. This ensures that the location (well, vial or tube) cannot be overfilled during fraction collection. This volume can be further reduced by defining a customized volume.

Additional check boxes in this section provide the opportunity to **Store** the **temperature signal** and the **UIB signal**.

Setting up a Fraction Collector Method

### **Fraction Preview**

To determine the appropriate fraction collection parameters the Agilent ChemStation provides a valuable tool that becomes accessible by pushing the button labelled Fraction Preview Tool (Figure 26) in the Peak Detectors section.

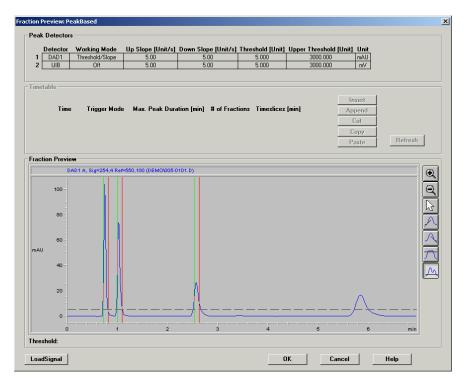


Figure 26 Fraction Preview dialog box

The Fraction Preview screen allows to test the fraction collection parameters against an example chromatogram. It can also be used to optimize the fraction collection parameters interactively. With the help of this tool values for up and down slope as well as for upper and lower threshold can easily be graphically specified. To load a chromatogram e.g. a pilot run click **Load Signal**. Parameters can now be changed either manually in the Detector Table and Timetable or graphically in the Fraction Preview screen. By pushing the desired buttons on the right hand side of **Fraction Preview** screen the chromatogram can be

zoomed, the values for up and down slope can be specified and the upper and lower threshold level can be set-up. The graphically specified values are automatically transferred to the Peak Detector Table.

## **Starting Your Run with Fraction Collection**

#### **Resetting the Current Fill Levels**

To protect the collected fractions against contaminations and to avoid overfilling of single positions the Agilent 1260 Infinity Fraction Collector stores current fill levels. It will block used positions, if no fraction start location has been specified.

If a new tray of the same type is inserted in the fraction collector, all fill levels can be reset (not just the well plate, vial or test tube). After re-installation of the tray a pop-up window as displayed in Figure 27 will be displayed. Click on **Yes** to reset all fill levels from the previous run.





### CAUTION

The window (Figure 27) to reset current fill levels is not displayed, if the well plate, the vial or the test tube is removed without removing the complete tray. Consequently contaminated fractions, unwanted overfill events or split fractions might be observed during the following LC run.

### **Assignment of Start Location for Fraction Collection**

The start location for fraction collection can either be assigned in the **Sample Info** (Figure 28) dialog box in the **RunControl** menu, in the **Sequence Parameters** (Figure 29) dialog box or in the **Sequence Table** in the **Sequence** menu. All fraction start locations are entered in the following formats.

#### For vials and test tubes:

fraction collector # - vessel number, e.g. 1-Vial 1

#### For well plates:

fraction collector # - plate number - row - column, e.g. 1-P1-A-01

with

fraction collector **#** = 1,2; plate number = P1-P4; row is A to H and column = 1-12 for 96-well plates.

#### NOTE

Note the orientation of the well plate. The starting position A1 is always in the back left corner of the plate.

Sample Info: Instrum	ent 1					X
Operator Name:	Bio Chemist		1			
-Data File						
C Prefix/Counter	Manual		Filen: DEFAU			
Su <u>b</u> directory:	ROTEOME					
Path: C:\HPCHE	M\1\DATA\					
Sample Paramete	rs					
		LOCation: Via		<u>/blank</u> run	if no entry)	
	Eractio	n Start Location: 1-P1	- <b>A-</b> 01			
Sample <u>N</u> ame:	Sample	Plate	ID:	1		
Sample <u>A</u> mount:	0	Multip	lier: 1			
ISTD Amount:	0	Dil <u>u</u> t	ion: 1			

Figure 28 Sample Info dialog box

All settings made in the Sequence Table overrule the settings made in the **Sequence Parameters** screen. In addition to the exact position for fraction collection start in the **Sequence Table** it is also possible to specify Next Plate and Next Location. In the former case fraction collection is started at the next free plate and in the latter case fraction collection is started at the next free location.

If a fraction start location is specified, the collector will start at this location, even if this vessel has been used during a previous run.

#### 2 Configuration and Operation of the Fraction Collector

**Starting Your Run with Fraction Collection** 

If no fraction start location has been specified, the fraction collection starts at the first empty vessel or location.

#### CAUTION

With multiple fraction collectors a fraction start location must be specified, e.g. 1-P1-A-01 or 2-Vial 10. Otherwise the fraction collection of the next run will start at the fraction collector, where the last run stopped. Never used the valve settings to define a start location.

quence Parameters: Instrument 1	
perator Name: Bio Chemist	
Data File	- Bar Code Reader
Auto C Prefix/Counter	□ Use in Sequence
Prefix: Counter:	On a bar code mismatch
SIG1 0001	C Inject anyway
Subdirectory: PROTEOME	@ Don't inject
Path: C:\HPCHEM\1\DATA\	
Part of methods to run	Shutdown
According to Runtime Checklist	Post-Sequence Cmd / Macro
Use Sequence Table Information	macro "SHUTDOWN.MAC", qo 👻
WaitTime: min (after loading a ne <del>w</del> method)	nRdy Timeout: min
Fraction Information Eraction Start Locatio : 1-P1-A-01	
equence Commen <u>t</u> :	
	A
OK	cel Help

Figure 29 Start Location in Sequence Parameters dialog box

To start a single run

- click on the Start button in the graphical user interface of the ChemStation.
- select Run Method from the RunControl menu.
- press F5

To start the sequence

- click on the Start button in the graphical user interface of the ChemStation.
- select Run Sequence from the RunControl menu.
- press F6

## **Online Tick Marks**

To display tick marks for fraction start and stop events in you Online Plot click on the **Change** button in your **Online Plot** window. Then check **Show fraction collection ticks** in the **Edit Signal Plot** window (Figure 30).

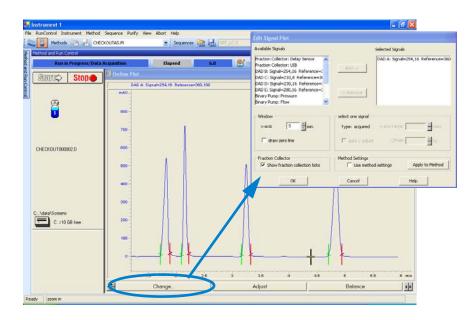


Figure 30 Online Tick Marks

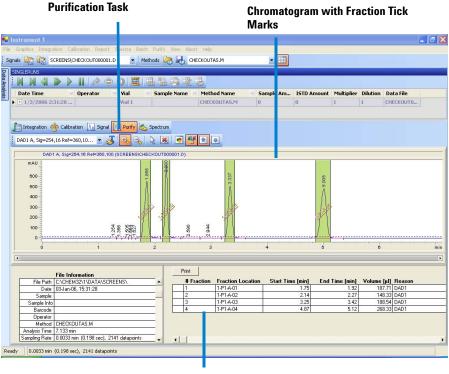
2 Configuration and Operation of the Fraction Collector Viewing Your Results

# **Viewing Your Results**

### **Data Analysis**

In order to display the tick marks for the collected fractions on the screen, click **Signal options** from the **Graphics** menu. Then choose **Separated** in the **Layout dropdown list**.

To review your chromatograms, file information and a fraction list, select the **Data Analysis** view and press **Fraction Task** as displayed in Figure 31.



Fraction table

Figure 31 Fraction Task button

In order to display the tick marks for the collected fractions on the screen, click **Signal options** from the **Graphics** menu. Then choose **Separated** in the Layout dropdown list.

### Report

In order to create reports with a fraction table and tick marks the Specify Report box the item Add Fraction Table and Tick Marks has to be checked.

pecify Report: Inst	rument 1	×
Destination		Quantitative Results
☐ Printer	I Screen	Calculate: Percent 💌
🗆 File	File Type	Based On: Area 💌
File Prefix	L.DIF L.CSV	Sorted By: Signal 💌
Report	LXLS LHTM	Signal Options
Style		
Report Style: S	Short 🗾 💌	
Add Chromat Report Layou	t For Uncalibrated Peaks —	Add Summed Peaks Table
- Chromatogram (	Jutput-	
C Portrait		-Size % of Page
CLandsc	ape	100 2
C Multi-Pe	age (Landscape)	Time: 100 - Response: 40 -
1	- Pages	
	OK Cancel	Help

Figure 32 Fraction collection report setup

2 Configuration and Operation of the Fraction Collector Special Applications

# **Special Applications**

### Pooling

We define Pooling as collecting fractions from multiple injections into the same fraction locations. In the Agilent Chemstation, there are two ways to initiate pooling:

- If you want to collect multiple injections from the one sample vial or sample well into the same fraction location, specify multiple injections in the Sequence Table.
- In case you want to collect multiple injections from the several sample vials or sample wells into the same fraction location, specify the same **Fract. Start** for multiple samples in the **Sequence Table**. An example is displayed in Figure 33 on page 78. In this sequence the Sample 1 will be injected from Plate 1 Position A1 of the Agilent 1260 Infinity High Performance Autosampler and fraction collection will start at Plate 1 Position A1 of the fraction collector.

For the next Sample 2, which is injected from a different location in the high performance autosampler, the fraction collection will start again at Plate 1 Position A1 of the fraction collector.

Line	Location	Sample Name	Dilution	Datafile	Inj Volume	Frac. Start
1	P1-A-01	Sample 1				1-P1-A-01
2	P1-A-02	Sample 2				1-P1-A-01
3	P1-A-03	Sample 3				1-P1-A-01
4						



#### CAUTION

If you specify multiple injections for pooling, the overfill protection is not activated. It is the user's responsibility to make sure that all fraction collection locations are large enough to completely collect all pooled fractions. If a fraction collection location is overfilled, an error message occurs and the pump is shut OFF.

### **Sample Recovery**

The Agilent 1260 Infinity Fraction Collectors offer different possibilities for sample recovery:

• The preferred recovery strategy is to install multiple fraction collectors in your LC systems and use the last of those fraction collectors for recovery. This recovery fraction collector can be selected in the Configuration dialog box as displayed in Figure 34. The fourth fraction collector in your systems will always be used for sample recovery. Also read "Configuring Multiple Fraction Collectors" on page 48.

Fraction Colle	ector 2 Configuration : Instrument 1		×
Trays Tray A:	40 Funnels	✓ Use for Sample Recovery	
Tray B:	40 Funnels	Tube Volume [ml]: 0.00	
Tray C:	40 Funnels	Tube Height [mm]: 0	

Figure 34 Sample Recovery Fraction Collector

- For the following tray configurations fixed recovery location will be assigned from the ChemStation. In order to disable the recovery the tray configuration has to be changed.
  - Standard tray for two well plates + 10 2ml x vials (G2258-60011) and half tray for 40 x 2 ml vials (G1313-44512).
     In this configuration the forty 2 ml vials on the half will automatically be used for recovery.
  - 2 Standard tray for two well plates + 10 x 2ml vials (G2258-60011) and half tray for 15x 6 ml vials (G1313-44513).
     In this configuration the fifteen 6 ml vials on the half will automatically be used for recovery.
  - 3 Standard tray for two well plates + 10 x 2ml vials (G2258-60011) and half tray with 40 funnels (G1364-84532).In this configuration the funnels on the half will automatically be used for recovery.
- The **Standard tray for two well plates + 10 funnels (G1364-84522)** allows two modes of operation.

#### 2 Configuration and Operation of the Fraction Collector

**Special Applications** 

- 1 If well-plates are configured in the fraction collector configuration the (checkbox **Plate 1** or **Plate 2** are selected), the 10 funnel locations will automatically be used for recovery.
- 2 If the checkbox **Plate 1** and **Plate 2** are not selected, the 10 funnel position can be used for regular fraction collection.

### CAUTION

With each start of a new sequence the recovery will start at the same positions. In order to avoid contamination the vessels that contain the recovery fractions have to be exchanged.

### CAUTION

The number of recovery locations automatically defines the maximum number of injections. When using the standard tray for two well plates and 10 funnels, only ten injections per sequence are executed.

### Semi-preparative Operation

The analytical-scale fraction collector G1364C can be modified for semi-preparative operation. This allows to use of external collection vessels through funnels at high flow rates above 10 ml/min. The maximum flow rate depends on the viscosity of the solvent.

In order to modify the analytical fraction collector the following parts have to be installed:

- 20 mm needle assembly (G1364-87202)
- preparative tubing kit (G1364-68711)
- internal tray preparative scale (G1364-63123)

The procedures to exchange these parts are described in "Replacing the Inlet/Waste Tubings" on page 105, "Replacing the Valve to Needle Tubing" on page 109, "Exchanging the Analytical Needle Assembly" on page 115, and "Exchanging the Internal Tray" on page 120.

In addition the wellplate adapter has to be removed for operation with the shorter needle.

In the ChemStation Software the new configuration has to be specified in order to consider the new volumes for the delay calibration. In the **Configuration** dialog box of the fraction collector select **More**. Then change the **Inner Capillary Diameter** to 0.8 mm as well as the **Needle Type** to semi-prep needle as displayed in Figure 35 on page 81. For further information on delay volumes and delay volume calibration see "Delay Volumes and Delay Calibration" on page 50.

Inner Capillary	Needle Type	
Diameter: 0.80 💌 mm	Needle Type:	semi-prep needle 💌 no needle micro needle
<u><u> </u></u>	Cancel	analytical needle semi-prep needle prep needle <u>II</u> eip

Figure 35 Fraction collector configuration parameters for semi-prep operation

### **Using High Test Tubes in the Analytical Scale Fraction Collector**

In the standard configuration the analytical-scale fraction collector G1364C the maximum test tube height is limited to 48 mm. This limitation can be extended to 75 mm if

- the standard 50 mm needle assembly (G1367-87200) is replaced by the 20 mm needle assembly (G1364-87202)
- the wellplate adapter is removed.

2 Configuration and Operation of the Fraction Collector Optimizing Fraction Collection

# **Optimizing Fraction Collection**

Time-based fraction collection	<ul> <li><i>Time slices</i> must have a length of at least 0.05 min.</li> <li>Set # of <i>Fractions</i> such that length of resulting fractions is at least 0.05 min.</li> </ul>
Peak-based fraction collection	<ul> <li>Set threshold and slope values such that length of fractions is at least 0.05 min.</li> <li>Unresolved peaks can be separated using appropriate threshold and slope values. If two unresolved peaks are to be collected as one fraction, collect based on threshold only.</li> <li>If the baseline of the chromatogram is below or above 0 mAU, this offset is not accounted for when triggering peaks using a threshold value. The threshold value is always added to 0 mAU.</li> </ul>

# **Limitations and How to Avoid Problems**

Rinse Fraction Collection Needle	If <i>Rinse Fraction Collection Needle</i> is set to <i>Between fraction collection</i> , at least 0.3 min are required to perform this task. When doing time-based fraction collection rinsing the needle is only possible between two time table entries, which must have a gap of at least 0.3 min. For peak-based fraction collection a time gap of also at least 0.3 min is required. If a new peak is detected during the rinse process, it is aborted and the needle moves back to the next free fraction position. Depending on flow rate and delay volume $V_{\rm D1}$ the beginning of this peak may be lost.
	If you have recovery positions in your fraction collector or if you are using one fraction collector for sample recovery in a multiple fraction collector configuration, the rinse function <b>between fraction collection</b> is ignored.
Needle Movement	The option <b>into location</b> under <b>Needle Movement</b> in the fraction collector configuration must only be used for capped 2 or 5 ml vials or well-plates. Using other or open vials with this command can lead to a <b>Movement failed</b> error.
Replacing fraction containers	When replacing filled tubes, vials or well-plates from the fraction collector make sure to remove and re-insert the complete tray. Otherwise the fraction collector will not recognize that the fraction containers were emptied.
Pooling	• When pooling fractions, overfill protection no longer exists. It is the user's responsibility to make sure that all fraction collection locations are large enough to completely collect all pooled fractions. If a fraction collection location is overfilled, an error message occurs and the pump is shut OFF.

2 Configuration and Operation of the Fraction Collector Check-out Procedures

# **Check-out Procedures**

The purpose of the check-out injection is the verification of the correct installation and configuration of the Agilent 1260 Infinity Purification System. In addition the checkout procedure can used for troubleshooting, if the Agilent 1260 Infinity Fraction Collector doesn't operate as expected.

The checkout procedure depends on the flow range (analytical scale AS or preparative scale PS) and on the peak trigger (UV-based or mass-based).

In the following sections the check-out procedures for the UV based trigger are described. For the mass-based system refer to the Agilent 1260 Infinity Purification System User Manual G2262-90011.

### Check-out Procedure for UV-triggered Fraction Collection AS

#### **Prerequisites**

- ✓ The Agilent 1260 Infinity LC System has been flushed thoroughly with the liquid phase as described in Table 14 and the lamp(s) of the UV detector have been switched on for at least 15 minutes to provide a stable baseline.
- ✓ The Agilent 1260 Infinity Fraction Collector has been installed and configured. The delay volume has been calibrated as described in "Performing a Delay Calibration with an UV Detector" on page 55.
- ✓ The 40-vial tray (G1313-44512) with at least four 2ml vials is installed.
- ✓ All wells are empty and the fill levels, which are stored on the module, have been reset. To verify this remove and insert the 40-vial tray. Then click on Yes in displayed dialog box.

Agilent 1260 Infinity modules	• standard or micro degasser
	<ul> <li>isocratic, quaternary or binary pump</li> </ul>
	<ul> <li>autosampler/ well plate sampler</li> <li>column compartment</li> </ul>
	<ul> <li>diode array detector, multi-wavelength detector or variable</li> </ul>
	wavelength detector with analytical flow cell
	<ul> <li>fraction collector analytical scale</li> </ul>
Column	ZORBAX Eclipse XDB-C18, 5 μm, 150 x 4.6mm (993967-906) or
	equivalent
Sample	Check-out sample (01080-68704)
Flow	0.8 ml/min
Stoptime	6 minutes
Solvent A	10% (HPLC grade water)
Solvent B	90% (HPLC grade acetonitrile)
Wavelength VWD	254 nm
Wavelength DAD/MWD	Signal A: 254/16nm, Reference: 360/80nm
Peakwidth (Responsetime)	> 0.1min (2s)
Injection volume	1 µl
Column temperature	40.0 <sup>0</sup> C
Fraction trigger mode	Peak-based
Max. peak duration	0.5 min
DAD/MWD working mode	Threshold/slope
Upslope	5 mAU/s
Downslope	10 mAU/s
Threshold/Upper threshold	10 mAU/ 3000 mAU

 Table 14
 Method Parameters for UV-triggered Fraction Collection Check-out AS

#### 2 Configuration and Operation of the Fraction Collector Check-out Procedures

#### **Expected Result**

Run a single injection under the conditions as described in Table 14. You should observe a chromatogram with four peaks as displayed in Figure 36 on page 86. Four fractions should have been collected in the fraction collector vials.

### NOTE

The retention times of the peaks could be different depending on the system configuration (e.g. flow cell type, capillary volumes, type and age of column).

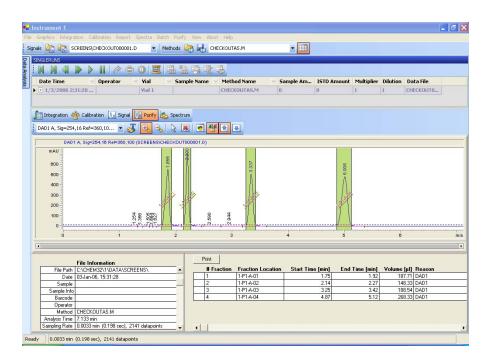


Figure 36 Chromatogram of the check-out injection with UV-based trigger

### **Check-out Procedure for UV-triggered Fraction Collection PS**

#### Prerequisites

- ✓ The Agilent 1260 Infinity LC System has been flushed thoroughly with the liquid phase as described in Table 14 and the lamp(s) of the UV detector have been switched on for at least 15 minutes to provide a stable baseline.
- ✓ The Agilent 1260 Infinity Fraction Collector has been installed and configured. The delay volume has been calibrated as described in "Performing a Delay Calibration with an UV Detector" on page 55.
- ✓ The 15-vial tray (G1313-44513) with at least four 6ml vials is installed.
- ✓ All wells are empty and the fill levels, which are stored on the module, have been reset. To verify this remove and insert the 15-vial tray. Then click on Yes in displayed dialog box.

Agilent 1260 Infinity modules	<ul> <li>preparative pump plus gradient extension (optional)</li> <li>preparative autosampler/ dual-loop autosampler</li> <li>diode array detector or multi-wavelength detector with preparative flow cell<sup>*</sup></li> <li>fraction collector preparative scale</li> </ul>
Column	ZORBAX SB-C18, 50 mm x 9.46mm (846975-202) or equivalent
Sample	Check-out sample (01080-68704)
Flow	10 ml/min
Stoptime	4 minutes
Solvent A	35% (HPLC grade water)
Solvent B	65% (HPLC grade acetonitrile)
Wavelength DAD/MWD	Signal A: 254/16nm, Reference: 360/80nm
Peakwidth (Responsetime)	> 0.1min (2s)
Injection volume	50 µl
Column temperature	ambient
Fraction trigger mode	Peak-based

**Table 15** Method Parameters for UV-triggered Fraction Collection Check-out PS

#### 2 Configuration and Operation of the Fraction Collector

**Check-out Procedures** 

Max. peak duration	0.5 min
DAD/MWD working mode	Threshold/slope
Upslope	5 mAU/s
Downslope	10 mAU/s
Threshold/Upper threshold	10 mAU/ 3000 mAU

#### Table 15 Method Parameters for UV-triggered Fraction Collection Check-out PS

\* The checkout method is tailored for the 3mm stainless steel flow cell G1315-60016. For other preparative flow cells some parameters (e.g. flow rate, injection volume, threshold/slope), should be adjusted.

#### **Expected Result**

Run a single injection under the conditions as described in Table 15. You should observe a chromatogram with four peaks as displayed in Figure 36 on page 86. Four fractions should have been collected in the fraction collector vials.

### NOTE

The retention times of the peaks could be different depending on the system configuration (e.g. flow cell type, capillary volumes, type and age of column).

# **Application Notes**

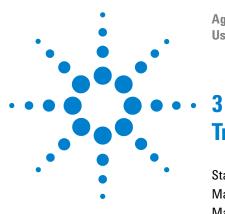
More information about the Agilent 1260 Infinity Fraction Collectors and Purification Systems are available from a series of application notes. Printed versions can be ordered from Agilent or pdf-files can be downloaded from the Agilent Website

http://www.chem.agilent.com

Title	Publication Number
An optimized system for analytical and preparative work	5988-9649EN
Recovery collection with the Agilent LC purification system	5988-9650EN
Solutions for preparative HPLC-Application Compendium	5988-9646EN
Innovative fraction collection with the Agilent LC purification platform	5988-9250EN
Automated fraction re-analysis - does it really make sense?	5988-8653EN
Injection of high-concentration samples with the Agilent LC purification system	5988-8654EN
Sophisticated peak-based fraction collection - working with up and down slope	5988-7895EN
Strategies for purification of compounds from non-baseline separated peaks	5988-7460EN
Method scale-up from analytical to preparative scale with the Agilent LC purification system PS	5988-6979EN
Peak-based fraction collection with the Agilent LC purification system AS - Influence of delay volume on recovery	5988-5747EN

#### Table 16 Selected Agilent Technologies Application Notes

#### 2 **Configuration and Operation of the Fraction Collector Application Notes**



Agilent 1260 Infinity Fraction Collectors User Manual

# **Troubleshooting and Test Functions**

Status Indicators 93 Maintenance Functions 95 Maintenance Functions 95 Transport Unit Self Alignment 97 Step Commands 99



### **Status Indicators**

The Fraction Collector is provided with two status indicators which indicate the operational state (prerun, not ready, run, and error states) of the instrument. The status indicators provide a quick visual check of the operation of the Fraction Collector (see "Status Indicators" on page 93).

### **Error Messages**

In the event of an electronic, mechanical or hydraulic failure, the instrument generates an error message in the user interface. For a detailed description of the failure, a list of probable causes of the problem, and a list of suggested actions refer to the *Agilent 1260 Infinity Fraction Collectors Service Manual G1364-90111*.

### **Maintenance Functions**

The maintenance functions position the transport unit and needle carrier assembly on certain positions for maintenance, homing, parking or delay calibration (see "Maintenance Functions" on page 95).

### **Transport Unit Self Alignment**

The transport unit alignment with the transport unit and the well-plate tray is required to compensate for larger deviations in positioning the needle carrier assembly.

The transport unit self alignment is required after disassembling the system or when you exchange the transport unit, the needle carrier assembly or the MTP main board. This function is in the **Diagnose** screen of the ChemStation or the Agilent Instant Pilot G4208A.

#### WARNING

The sample transport self alignment requires one of the 4-well-plate trays (G1364-84501), but well plates MUST NOT be installed!

# **Status Indicators**

Two status indicators are located on the front of the Fraction Collector. The lower left indicates the power supply status, the upper right indicates the Fraction Collector status.

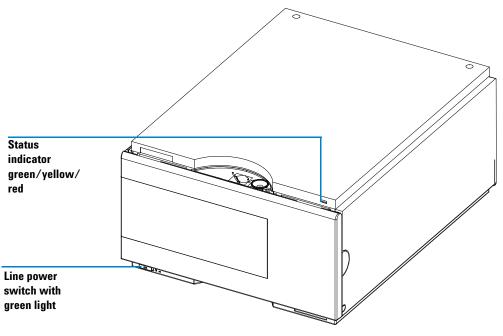


Figure 37 Location of Status Indicators

### **Power Supply Indicator**

The power supply indicator is integrated into the main power switch. When the indicator is illuminated (*green*) the power is ON.

### **Instrument Status Indicator**

The instrument status indicator indicates one of four possible instrument conditions:

- When the status indicator is OFF (and power switch light is on), the instrument is in a *prerun* condition, and is ready to begin an analysis.
- A *green* status indicator indicates the instrument is performing an analysis (*run* mode).
- A *yellow* status indicator indicates a *not-ready* condition. The instrument is in a not-ready state when it is waiting for a specific condition to be reached or completed (for example, front door not closed), or while a self-test procedure is running.
- An *error* condition is indicated when the status indicator is *red*. An error condition indicates the instrument has detected an internal problem which affects correct operation of the instrument. Usually, an error condition requires attention (for example, leak, defective internal components). An error condition always interrupts the analysis.

# **Maintenance Functions**

Some maintenance procedures require the needle arm, and needle carrier to be moved to specific positions to enable easy access to components. The maintenance functions move these assemblies into the appropriate maintenance position. In the ChemStation the fraction collector maintenance positions can be selected from the **Maintenance menu** in the **Diagnosis** view (see Figure 38). In the Agilent Instant Pilot G4208A the functions can be selected in the **Test** screens of the fraction collector.

Fraction collector Maintenance Positions 🛛 🛛 🕅	
Change Parts	
	itart
I	End
Move Arm Home	Park Arm for Transport
Home	Park Arm
No tray	Close

Figure 38 Fraction Collector Maintenance Positions dialog box

#### **Change Parts.**

If you click **Start** the transport unit will move upwards, the needle carrier assembly will move to the front center and then turn OFF the theta motor to allow free rotation of the theta arm. This position enables easy access to the transport unit to change:

#### **3** Troubleshooting and Test Functions

**Maintenance Functions** 

- The needle assembly
- The needle carrier assembly
- The valve to needle tubing.
- The diverter valve.

After the maintenance or repair task has been finished, click **End** to move the transport assembly to the **Home** position

#### **Home Position**

This maintenance function moves the arm up and to the right rear for better access and exchange of the trays.

#### Park Arm

This maintenance position moves the arm to the park position at the upper rear left side of the tray for transporting or shipping the Fraction Collector.

# **Transport Unit Self Alignment**

The transport unit alignment with the transport unit and the well-plate tray is required to compensate for larger deviations in positioning the needle carrier assembly.

The transport unit self alignment is required after disassembling the system or when you exchange the transport unit, the needle carrier assembly or the MTP main board.

This function is in the **Diagnose** screen of the ChemStation or the Agilent Instant Pilot G4208A.

# WARNING The sample transport self alignment requires one of the 4-well-plate trays (G1364-84501), but well plates MUST NOT be installed.

If the Transport Unit Self Alignment is started with well plates on the tray, the alignment procedure is aborted WITHOUT error message.

#### When is a Transport Unit Self Alignment Necessary?

The sample transport self alignment is required after disassembling the module or when you exchange:

- The transport unit.
- The needle/capillary carrier assembly.
- The MTP main board.

#### How to perform a Transport Unit Self Alignment?

Steps	Comments	
	For details see: "Forced Cold Start" in the Fraction Collectors Service Manual.	
2 Install the 4-well-plate tray (G1364-84501)	IMPORTANT: Remove all plates!	

#### How to perform a Transport Unit Self Alignment?

Steps		Comments	
3	Ensure that the wellplate adapter is correctly assembled	On a G1364B preparative-scale fraction collector install the delay calibration adapter G1364-87301	
4	Select the <b>Maintenance</b> menu in the <b>Diagnosis</b> view of the Agilent ChemStation.		
5	In the menu choose <b>Fraction Collector &gt;</b> <b>Transport Alignment</b> to start the automated procedure.	The Transport Alignment Procedure takes approximately 10-15 minutes	
6	Set the 8-bit configuration switch to the default setting.	See "8-bit configuration switch" in the Fraction Collectors Service Manual.	

### NOTE

If the Transport Unit Self Alignment is started with well plates on the tray, the alignment procedure is aborted without any error message.

# **Step Commands**

Some movements of the fraction collection sequence can be done under **Manual control**. This is useful during troubleshooting where close observation of each of the fraction collection step is required to confirm a specific failure mode or verify successful completion of a repair.

Each step command actually consists of a series of individual commands which move the Fraction Collector components to predefined positions enabling the specific step to be done.

In the ChemStation the step commands can be selected from the **"Test Selection Box**" (see Figure 39) in the **Diagnosis** display. In the Agilent Instant Pilot G4208A the step commands can be accessed from the pull-down menu in the Fraction Collector **"Test**".

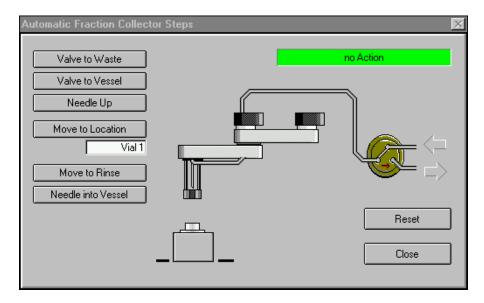


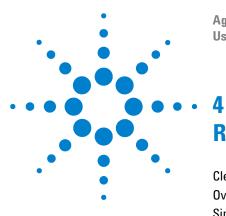
Figure 39 Fraction Collector Step Commands

Step	Action	Comments
Needle Up	Lifts the needle arm to the upper position.	Command also switches the diverter valve to waste if it is not already in that position.
Needle into vessel	Lowers the needle into the specified vessel.	only analytical-scale G1364C
Needle to rinse / flush port	Moves the needle to the rinse / flush port.	only analytical scale G1364C
Switch valve to needle	Switches the diverter valve to the needle.	
Switch valve to waste	Switches the diverter valve from needle to waste.	

 Table 17
 Step Commands

## Troubleshooting

If the Fraction Collector is unable to perform a specific step due to a hardware failure, an error message is generated. You can use the step commands to perform a fraction collection sequence, and observe how the Fraction Collector responds to each command.



Agilent 1260 Infinity Fraction Collectors User Manual

# **Repairing the Fraction Collector**

Cleaning the Fraction Collector 102 Overview of Main Repair Procedures 103 Simple Repairs 104



Agilent Technologies

# Introduction into Repairing the Fraction Collector

### **Simple Repairs**

The Fraction Collector is designed for easy repair. The most frequent repairs such as changing a needle assembly or tubings can be done from the front of the instrument with the instrument in place in the system stack. These repairs are described in "Simple Repairs" on page 104.

### WARNING

When opening capillary or tube fittings solvents may leak out. Please observe appropriate safety procedures (for example, goggles, safety gloves and protective clothing) as described in the material handling and safety data sheet supplied by the solvent vendor, especially when toxic or hazardous solvents are used.

### WARNING

To avoid personal injury, keep fingers away from the needle area during Fraction Collector operation. Do not bend the safety flap away from its position, or attempt to insert or remove a vial from the gripper when the gripper is positioned below the needle.

### WARNING

Regularly inspect the inlet / waste tubing assembly, and the valve to needle tubing and exchange them if they are worn out or show visible signs of damage.

### **Cleaning the Fraction Collector**

The Fraction Collector covers should be kept clean. Cleaning should be done with a soft cloth slightly dampened with water or a solution of water and a mild detergent. Do not use an excessively damp cloth from which liquid could drip into the Fraction Collector.

#### WARNING

Do not let liquid drip into the Fraction Collector. It could cause a shock hazard or damage to the Fraction Collector.

#### **Repairing the Fraction Collector** 4

**Overview of Main Repair Procedures** 

# **Overview of Main Repair Procedures**

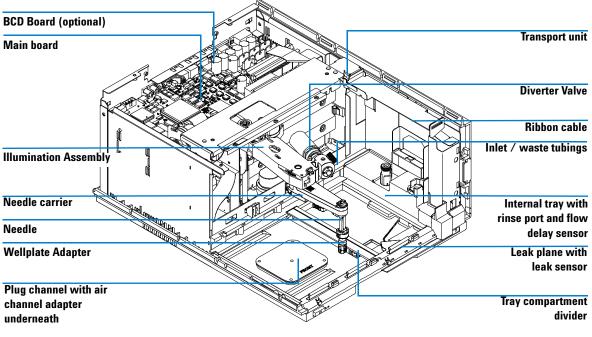


Figure 40 Main Assemblies (Example displays the analytical-scale module G1364C. The preparative-scale fraction collector G1364B contains a different needle)

# **Simple Repairs**

The procedures described in this section can be done with the Fraction Collector in place in the stack. These procedures can be done on a more frequent basis.

 Table 18
 Simple Repair Procedures

Procedure	Typical Frequency	Notes
Replacing the Inlet / waste tubings	When worn out, when showing visual signs of damage, typically once per year.	See "Replacing the Inlet/Waste Tubings" on page 105
Replacing the valve to needle tubing	When worn out, when showing visual signs of damage, typically once per year	See "Replacing the Valve to Needle Tubing" on page 109
Exchanging the <b>preparative</b> needle assembly	When needle shows indication of damage or blockage	See "Exchanging the Preparative Needle Assembly" on page 113
Exchanging the <b>analytical</b> needle assembly	When needle shows indication of damage or blockage Or when using the short needle assembly for operation with high test tubes (>45mm)	See "Exchanging the Analytical Needle Assembly" on page 115
Exchanging the diverter valve	When defective (internal / external leak, valve not switching any more)	See "Exchanging the Diverter Valve" on page 117
Exchanging the internal tray	When flow delay sensor defective	See "Exchanging the Internal Tray" on page 120
Repairing or exchanging a funnel of the internal tray or funnel tray	When defective (leaky, blocked or contaminated)	See "Repairing or Exchanging a Funnel of the Internal Tray" on page 122

# **Replacing the Inlet/Waste Tubings**

Frequency	When contaminated, worn out or visibly damaged Typically once every year
Tools required	None
Parts Required	Inlet / waste tubing assembly in tubing kit <b>preparative scale 0.8 mm ID,</b> PN G1364-68711, <b>analytical scale 0.25 mm ID</b> , PN G1364-68712 or <b>analytical-scale 0.15 mm ID</b> , G1364-68723



To avoid personal injury, keep fingers away from the needle area during Fraction Collector operation.

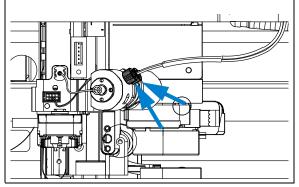
### WARNING

Follow the described installation procedure exactly to maximize the lifetime of the inlet / waste tubings and to avoid potential spills or fraction losses. Regularly inspect the tubings and exchange them if they are worn out or show visible signs of damage.

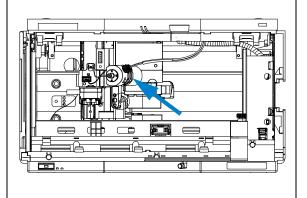
#### Before beginning this procedure:

- Position the transport unit of the Fraction Collector in the "Home Position" (see "Maintenance Functions" on page 95).
- Remove all installed trays from the tray base.
- Position the transport unit of the Fraction Collector in the "**Change Parts Position**" (see "Maintenance Functions" on page 95).
- Turn OFF the instrument.
- Remove the rear end of the Fraction Collector's waste tubing from the waste container, unscrew the front end of the Fraction Collector's inlet tubing from the flow cell of the detector.

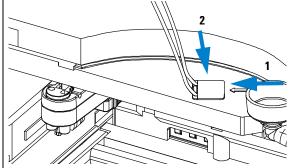
**2** Unscrew the 2 finger-tight fittings of the inlet / waste tubing assembly at the diverter valve.



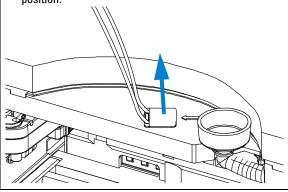
**1** Locate the diverter valve with the finger-tight fittings of the inlet / waste tubing assembly (the figure shows the open Fraction Collector seen from the front).



**3** Unplug the inlet / waste tubing assembly from the bolt carrier (located on the top center of the front cover) by moving the snapper as indicated by the arrows. Slide the tubings out to the bottom.

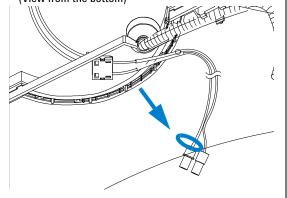


**4** Install the new inlet / waste tubing assembly into the bolt carrier as shown below. Slide in the long ends of the tubings from bottom to top and let the snapper click into position.

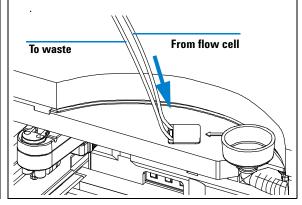


6 Connect the finger-tight fittings of the inlet / waste tubing assembly to the ports of the diverter valve. IMPORTANT: The tubings must not be bent up- or downwards. The cables must not be twisted.

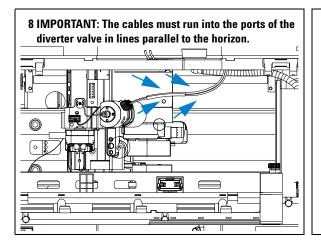
(View from the bottom)



**5** The rear tubing (with the label with the arrow) must be connected to the flow cell of the detector. The front tubing must be inserted into the waste container



7 A color coded ring on one of the tubings and the valve body indicates, which cable belongs to which port. IMPORTANT: It is absolutely vital to connect these tubings as described, in order to maximize their lifetime and operating security.



#### On completion of this procedure:

- Re-install the tray(s) in the tray base.
- Start the instrument.
- Close the front cover.

## **Replacing the Valve to Needle Tubing**

Frequency	When contaminated, worn out or visibly damaged Typically once every year
Tools required	Wrench, open end, 4mm, PN 8710-1534 (supplied in accessory kit) Wrench, open end, 1/4 – 5/16 inch, PN 8710-0510 (supplied in accessory kit)
Parts Required	Valve to needle tubing assembly included in tubing kit <b>preparative scale 0.8</b> mm ID, PN G1364-68711, analytical scale 0.25 mm ID, PN G1364-68712 or analytical scale 0.15 mm ID, G1364-68723

### WARNING

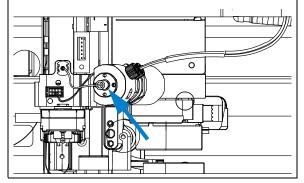
To avoid personal injury, keep fingers away from the needle area during Fraction Collector operation.

### WARNING

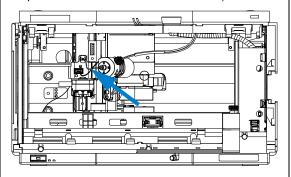
Follow the described installation procedure exactly to maximize the lifetime of the valve to needle tubing and to avoid potential spills or fraction losses. Regularly inspect the tubings and exchange them if they are worn out or show visible signs of damage.

#### Before beginning this procedure:

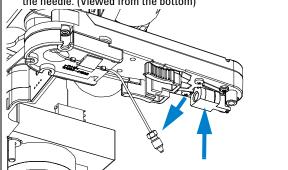
- Position the transport unit of the Fraction Collector in the "**Home Position**" (see "Maintenance Functions" on page 95).
- Remove all installed trays from the tray base.
- Position the transport unit of the Fraction Collector in the "**Change Parts Position**" (see "Maintenance Functions" on page 95) and turn off the instrument.
- It might be more convenient to remove the needle from its carrier before unscrewing the needle tubing.
- **2** Unscrew the finger-tight fitting of the valve to needle tubing assembly at the diverter valve.

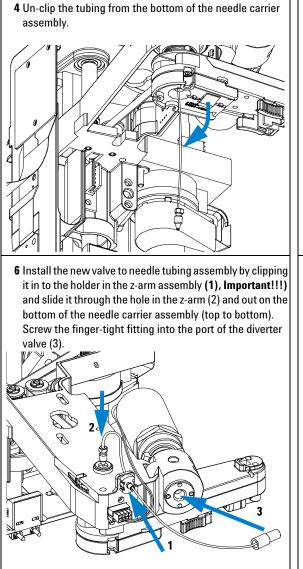


1 Locate the diverter valve with the finger-tight fittings of the valve to needle tubing assembly (the figure shows the open Fraction Collector seen from the front).

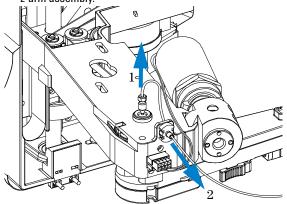


**3** Using the 4 mm wrench and the 5/16" wrench for counter-holding unscrew the valve to needle tubing from the needle. (Viewed from the bottom)

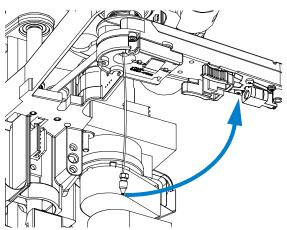




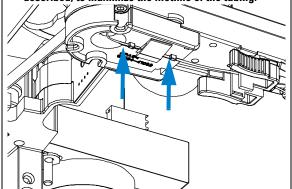
**5** Slide the tubing through the hole in the needle carrier assembly (from bottom to top) and out of the holder in the z-arm assembly.



7 Using the 4 mm wrench and the 5/16" wrench for counter-holding connect the valve to needle tubing to the needle. (Viewed from the bottom)



8 IMPORTANT: After fixing the screw clip the tubing into the guide on the bottom of the needle carrier assembly. It is absolutely vital that the tubing is installed as described, to maximize the lifetime of the tubing.



#### On completion of this procedure:

- Re-install the needle to the needle carrier assembly, if you previously removed it. Make sure to slide the needle all the way to the front of the needle carrier assembly (clicks into position).
- Re-install the tray(s) in the tray base.
- Start the instrument.
- Close the front cover.

## **Exchanging the Preparative Needle Assembly**

Frequency	When the needle is leaky or visibly damaged When the needle is blocked or contaminated
Tools required	Wrench, open end, 4mm, PN 8710-1534 (supplied in accessory kit) Wrench, open end, 1/4 – 5/16 inch, PN 8710-0510 (supplied in accessory kit)
Parts required	Preparative needle assembly, G1364-87201

### WARNING

To avoid personal injury, keep fingers away from the needle area during Fraction Collector operation.

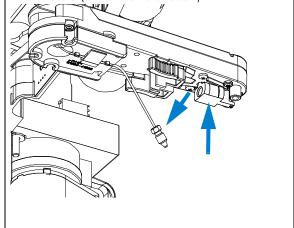
### CAUTION

Regularly inspect the inlet / waste tubing assembly and the valve to needle tubing and exchange them if they are worn out or show visible signs of damage.

### Before beginning this procedure:

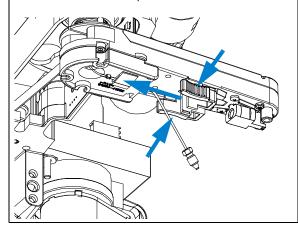
- Position the transport unit of the Fraction Collector in the "Home Position" (see "Maintenance Functions" on page 95).
- Remove all installed trays from the tray base.
- Position the transport unit of the Fraction Collector in the "Change Parts Position" (see "Maintenance Functions" on page 95).
- Turn OFF the instrument.
- It might be more convenient to remove the needle from its carrier before unscrewing the needle tubing.

**1** Using the 4 mm wrench and the 5/16" wrench for counter-holding unscrew the valve to needle tubing from the needle. (Viewed from the bottom)

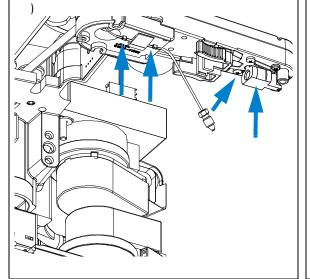


**Simple Repairs** 

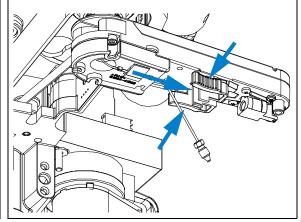
2 Holding the needle assembly between your thumb and forefinger, slide out the assembly towards the rear of the needle carrier assembly.



4 Using the 4 mm wrench and the 5/16" wrench for counter-holding connect the valve to needle tubing to the needle. (Viewed from the bottom



**3** Insert the new needle assembly into the holder of the needle carrier assembly. Make sure to push it all the way to the front.



#### On completion of this procedure:

- Re-install the needle to the needle carrier assembly, if you previously removed it. Make sure to slide the needle all the way to the front of the needle carrier assembly (clicks into position).
- IMPORTANT: After fixing the needle in the needle carrier clip the tubing into the guide on the bottom of the needle carrier assembly in case it slipped out of this guide (see 2 left arrows on the figure to the left). It is absolutely vital that the tubing is

It is absolutely vital that the tubing is installed as described, to maximize the lifetime of the tubing.

- Re-install the tray(s) in the tray base.
- Start the instrument.
- Close the front cover.

## **Exchanging the Analytical Needle Assembly**

Frequency	When the needle is visibly damaged When the needle is blocked or contaminated
Tools required	Wrench, open end, 4mm, PN 8710-1534 (supplied in accessory kit) Wrench, open end, 1/4 – 5/16 inch, PN 8710-0510 (supplied in accessory kit)
Parts required	Analytical needle assembly, G1367-87200

### WARNING

To avoid personal injury, keep fingers away from the needle area during Fraction Collector operation.

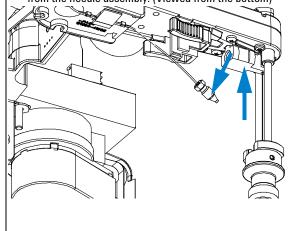
### CAUTION

Regularly inspect the inlet / waste tubing assembly and the valve to needle tubing and exchange them if they are worn out or show visible signs of damage.

### Before beginning this procedure:

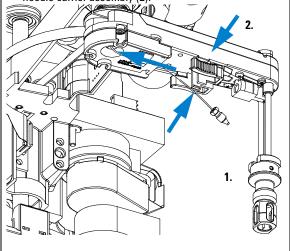
- Position the transport unit of the Fraction Collector in the "Home Position" (see "Maintenance Functions" on page 95).
- Remove all installed trays from the tray base.
- Position the transport unit of the Fraction Collector in the "Change Parts Position" (see "Maintenance Functions" on page 95).
- Turn OFF the instrument.
- It might be more convenient to remove the needle from its carrier before unscrewing the needle tubing.

**1** Use the 4 mm wrench and the 5/16" wrench for counter-holding to unscrew the valve-to-needle tubing from the needle assembly. (Viewed from the bottom)

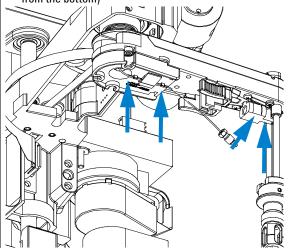


**Simple Repairs** 

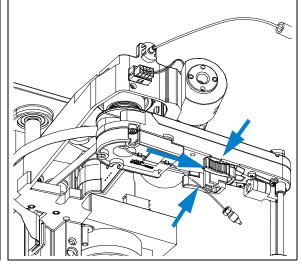
2 Remove the wellplate adapter by turning it clockwise (1). Hold the needle assembly between your thumb and forefinger, slide out the assembly towards the rear of the needle carrier assembly (2).



4 Use the 4 mm wrench and the 5/16" wrench for counter-holding to connect the valve to needle tubing to the needle. Reassembly the wellplate adapter. (Viewed from the bottom)



**3** Insert the new needle assembly into the holder of the needle carrier assembly. Make sure to push it all the way to the front. Reassemble the wellplate adapter.



On completion of this procedure:

• IMPORTANT: After fixing the needle in the needle carrier clip the tubing into the guide on the bottom of the needle carrier assembly in case it slipped out of this guide (see 2 left arrows on the figure to the left).

It is absolutely vital that the tubing is installed as described, to maximize the lifetime of the tubing.

- Re-install the tray(s) in the tray base.
- Start the instrument.
- Close the front cover.

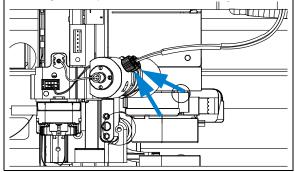
### Repairing the Fraction Collector 4 Simple Repairs

## **Exchanging the Diverter Valve**

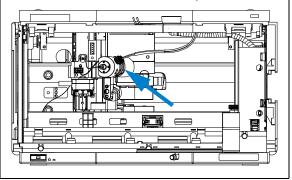
Frequency	When leaky or defective
Tools required	Hex key 2.0 mm, PN 8710-2438 (supplied in accessory kit)
Parts required	Diverter Valve, PN G1364-61901

#### Before beginning this procedure:

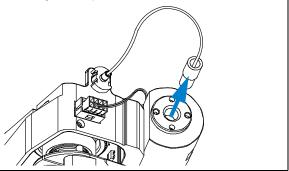
- Position the transport unit of the Fraction Collector in the "Home Position" (see "Maintenance Functions" on page 95).
- Remove all installed trays from the tray base.
- Position the transport unit of the Fraction Collector in the "Change Parts Position" (see "Maintenance Functions" on page 95).
- Turn OFF the instrument.
- **2** Unscrew the 2 finger tight fittings of the inlet / waste tubing assembly at the diverter valve.



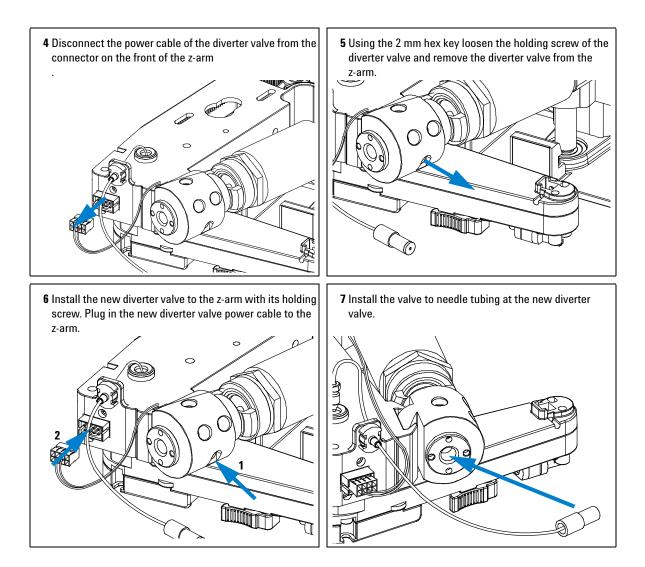
1 Locate the diverter valve with the finger tight fittings of the inlet / waste tubing assembly (the figure shows the open Fraction Collector seen from the front).

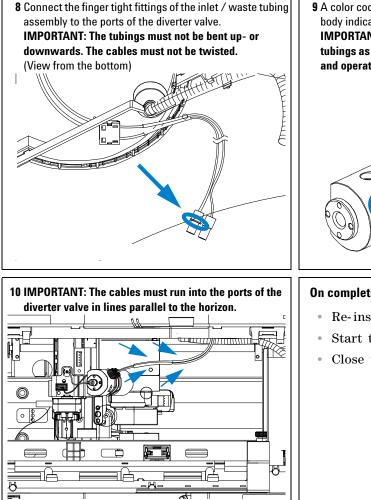


**3** Unscrew the finger tight fitting of the valve to needle tubing assembly at the diverter valve.



**Simple Repairs** 





9 A color coded ring on one of the tubings and the valve body indicates, which cable belongs to which port. IMPORTANT: It is absolutely vital to connect these tubings as described, in order to maximize their lifetime and operating security.

#### On completion of this procedure:

- Re-install the tray(s) in the tray base.
- Start the instrument.
- Close the front cover.

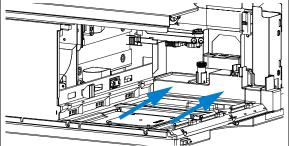
## **Exchanging the Internal Tray**

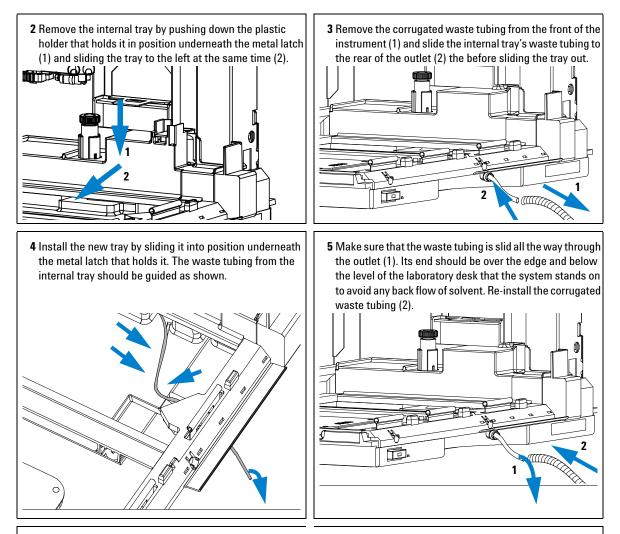
Frequency	When defective
Tools required	None
Parts required	Internal tray <b>analytical scale</b> , (G1364-63124) Internal tray <b>preparative scale</b> ,(G1364-63123)

### Before beginning this procedure:

- Position the transport unit of the Fraction Collector in the "**Home Position**" (see "Maintenance Functions" on page 95).
- Remove all installed trays from the tray base.
- Turn OFF the instrument.

1 Locate the internal tray assembly with the rinse funnel and flow delay sensor in the bottom of the right front corner of the instrument.





#### On completion of this procedure:

- Re-install the tray(s) in the tray base.
- Start the instrument.
- Close the front cover.

### 4 Repairing the Fraction Collector

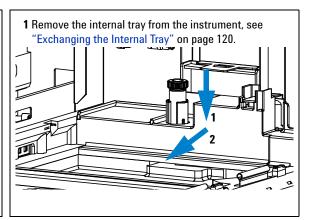
**Simple Repairs** 

## **Repairing or Exchanging a Funnel of the Internal Tray**

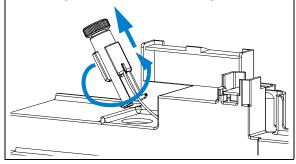
Frequency	When leaky or contaminated
Tools required	None
Parts required	Funnel assembly, waste tubing assembly, seals (for part numbers see "Internal Tray Assembly" on page 139)

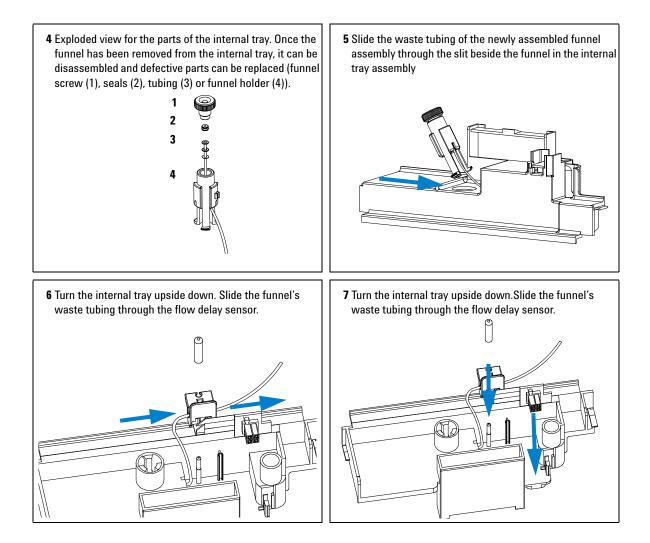
#### Before beginning this procedure:

- Position the transport unit of the Fraction Collector in the "**Home Position**" (see "Maintenance Functions" on page 95) and remove all installed trays from the tray base.
- Turn OFF the instrument.
- The procedure is almost identical for both, the preparative and the analytical scale internal tray, but it requires different parts.
- 2 Turn the internal tray upside down and remove the funnel's waste tubing through the flow delay sensor.

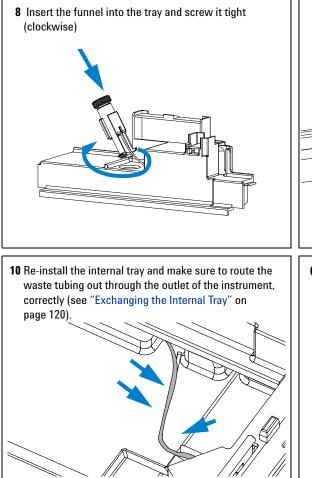


**3** Remove the funnel assembly from the internal tray by screwing it counter clockwise and lifting it out.

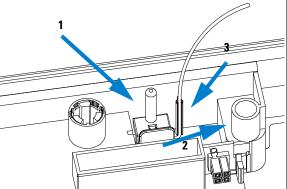




**Simple Repairs** 



**9** Re-install the plug that holds the flow delay sensor (1), pull the waste tubing tight through the flow delay sensor (2) and mount the waste tubing (3).



### On completion of this procedure:

- Re-install the tray(s) in the tray base.
- Start the instrument.
- Close the front cover.



Agilent 1260 Infinity Fraction Collectors User Manual

# **Parts and Materials**

Supported Trays for Fraction Collectors 126 List of Recommended Test Tubes 128 List of Recommended Vials and Caps 129 List of Recommended Plates and Closing Mats 132 Transport Unit Assembly (Preparative Scale) 134 Transport Unit Assembly (Analytical Scale) 135 Needle Assemblies 136 Diverter-Valve Assembly 137 Tubing Kits 138 Internal Tray Assembly 139 Fraction Collector Accessory Kit 140



Agilent Technologies

**Supported Trays for Fraction Collectors** 

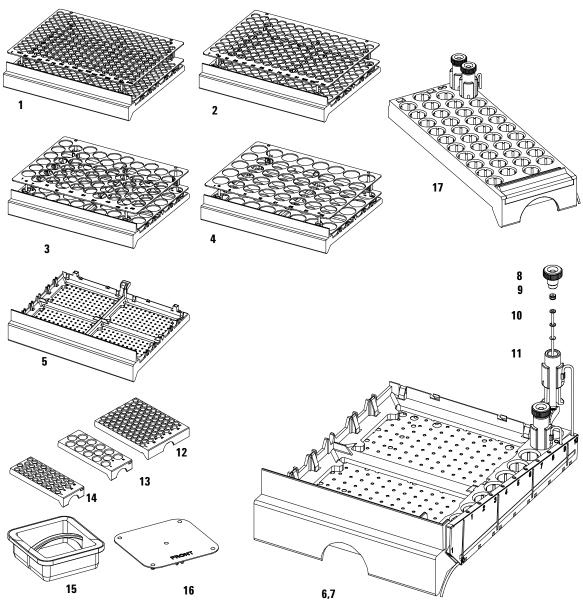
# **Supported Trays for Fraction Collectors**

ltem	Description	Part Number
1	Full tray for 215 test tubes, 12x100 mm	G1364-84516
2	Full tray for 126 test tubes, 16x100 mm	G1364-84525
3	Full tray for 60 round bottom tubes, 25x100 mm	G1364-84524
4	Full tray for 40 round bottom tubes, 30x100 mm	G1364-84523
5	Full tray for 4 well plates	G1364-84521
6	Std. tray for 2 well plates + 10 2ml vials	G2258-60011
7	Std. tray for 2 well plates + 10 collecting funnels	G1364-84522
8	Funnel for item 7	5022-2200
9	Funnel seal kit for item 7 (pack. of 10)	G1364-68730
10	Tubing kit 10T for item 7 (pack of 10)	G1364-86707
11	Funnel coupler for item 7	G1364-43201
12	Std. tray for 100 x 2 ml vials Std. tray for 100 x 2 ml vials, thermostattable	G1313-44510 G1329-60011
13	Half tray for 15 x 6 ml vials	G1313-44513
14	Half tray for 40 x 2 ml vials	G1313-44512
15	Adapter air channel (installed underneath plug channel, if the fraction collector is used with the thermostat)	G1329-43200
16	Plug channel	G1364-47200
17	Half Tray for 40 Funnels	G1364-84532

### Table 19 Trays for the Fraction Collector

NOTE

Only one type of well-plates can be used at a time in one tray.



The items in this figure are displayed in different scaling factors!

Figure 41 Trays

**Agilent 1260 Infinity Fraction Collectors User Manual** 

### 5 Parts and Materials

**List of Recommended Test Tubes** 

# **List of Recommended Test Tubes**

outer diameter	height	Volume	recommended tray type	Part Number
30 mm	100 mm	45 ml	G1364-84503 (40 tubes)	5042-6459 (100/pk)
30 mm	48 mm	20 ml;	G1364-84503 (40 tubes)	5042-6458 (100/pk)
25 mm	100 mm	35 ml	G1364-84504 (60 tubes)	5042-6470 (100/pk)
16 mm	100 mm	19 ml	G1364-84505 (126 tubes)	5022-6532 (250/pk)
16 mm	48 mm	9 ml	G1364-84505 (126 tubes)	5022-6533 (100/pk)
12 mm	100 mm	11 ml	G1364-84506 (215 tubes)	5022-6431 (250/pk)
12 mm	48 mm	5 ml	G1364-84506 (215 tubes)	5022-6435 (100/pk)

Table 20Round Bottom Test Tubes

## **List of Recommended Vials and Caps**

Description	Volume (ml)	IOO/Pack	l000/Pack	l00/Pack (silanized)
Clear glass	2	5181-3375	5183-4491	
Clear glass, write-on spot	2	5182-0543	5183-4492	5183-4494
Amber glass, write-on spot	2	5182-3376	5183-4493	5183-4495

 Table 21
 Crimp Top Vials (Caps for Use with the Analytical Scale Fraction Collector, only!)

 Table 22
 SnapTop Vials (Caps for Use with the Analytical Scale Fraction Collector, only!)

Description	Volume (ml)	l00/Pack	l000/Pack	IOO/Pack (silanized)
Clear glass	2	5182-0544	5183-4504	5183-4507
Clear glass, write-on spot	2	5182-0546	5183-4505	5183-4508
Amber glass, write-on spot	2	5182-0545	5183-4506	5183-4509

 Table 23
 Screw Top Vials (Caps for Use with the Analytical Scale Fraction Collector, only!)

Description	Volume (ml)	100/Pack	l000/Pack	l00/Pack (silanized)
Clear glass	2	5182-0714	5183-2067	5183-2070

#### **5** Parts and Materials

**List of Recommended Vials and Caps** 

Description	Volume (ml)	IOO/Pack	l000/Pack	l00/Pack (silanized)
Clear glass, write-on spot	2	5182-0715	5183-2068	5183-2071
Amber glass, write-on spot	2	5182-0716	5183-2069	5183-2072

 
 Table 23
 Screw Top Vials (Caps for Use with the Analytical Scale Fraction Collector, only!) (continued)

 Table 24
 Crimp Caps (Caps for Use with the Analytical Scale Fraction Collector, only!)

Septa	100/Pack
Clear PTFE/red rubber	5181-1210
Clear PTFE/red rubber	5183-4498 (1000/Pack)
Clear PTFE/red rubber	5181-1215
Clear PTFE/red rubber	5181-1216
Clear PTFE/red rubber	5181-1217
	Clear PTFE/red rubber Clear PTFE/red rubber Clear PTFE/red rubber Clear PTFE/red rubber

 Table 25
 Snap Caps (Caps for Use with the Analytical Scale Fraction Collector, only!)

Description	Septa	100/Pack
Clear polypropylene	Clear PTFE/red rubber	5182-0550
Blue polypropylene	Clear PTFE/red rubber	5182-3458
Green polypropylene	Clear PTFE/red rubber	5182-3457
Red polypropylene	Clear PTFE/red rubber	5182-3459

Description	Septa	100/Pack
Blue polypropylene	Clear PTFE/red rubber	5182-0717
Green polypropylene	Clear PTFE/red rubber	5182-0718
Red polypropylene	Clear PTFE/red rubber	5182-0719
Blue polypropylene	Clear PTFE/silicone	5182-0720
Green polypropylene	Clear PTFE/silicone	5182-0721
Red polypropylene	Clear PTFE/silicone	5182-0722

 Table 26
 Screw Caps (Caps for Use with the Analytical Scale Fraction Collector, only!)

**List of Recommended Plates and Closing Mats** 

## **List of Recommended Plates and Closing Mats**

Description	Volume (ml)	Package	Part Number
96 polypropylene well-plate	0.5	10	5042-1386
96 polypropylene well-plate	0.5	120	5042-1385
96 polypropylene deep well pate	1.0	50	5042-6454
96 polypropylene deep well-plate with glass inserts, caps and septa pre assembled <sup>*</sup>	0.35	1	5065-4402
glass inserts for 5065-4402 <sup>*</sup>	0.35	1000	5188-5321
384 polypropylene well-plate	0.1	30	5042-1388
96 polypropylene conical-well plate <sup>*</sup>	0.18	25	5042-8502
54 x 2ml vial plate <sup>*</sup>	1.5	6	G2255-68700
15 x 6ml vial plate	4.5	1	5022-6539
Plate for 27 self lock Eppendorf tubes	0.5, 1.5, 2	1	5022-6538
Silicon Closing mats for 96 well-plate		50	5042-1389

 
 Table 27
 Recommended Plates and Closing Mats (Std. Well Plates and Closing Mats for Use with the Analytical Scale Fraction Collector, only!)

\* For use with the **analytical scale** Fraction Collector, only!

### NOTE

Only one type of well-plates can be used at a time in one tray.

## WARNING

If you are using flammable solvents, remove the plates from the Fraction Collector after turning it OFF. You avoid the risk of building explosive gas mixtures in the instrument.

## WARNING

If you are using flammable solvents, cover the plates. This action avoids the risk of building explosive gas mixtures in the instrument.

WARNING

Closing mats with adhesive can give some contamination in the system. The adhesive is soluble in most of the solvents used in HPLC.

### WARNING

In general do not use closing mats with adhesive. The Fraction Collector has no prepunch needle, therefore the adhesive will clog the needle after several injections.

### 5 Parts and Materials

Transport Unit Assembly (Preparative Scale)

# **Transport Unit Assembly (Preparative Scale)**

ltem	Description	Part Number
1	Transport Unit Assembly ( <b>Preparative Scale)</b> , includes items 2 and 4	G1364-60018
2a, b	Tubing - diverter valve to needle, 0.8 mm ID	G1364-86711
3	Diverter valve assembly	G1364-61901
4	Needle carrier assembly preparative scale	G1364-60011
5	Needle assembly preparative scale	G1364-87201
6	Delay Calibration Adapter (not shown)	G1364-87301

### Table 28 Transport Unit Assembly (Preparative Scale)

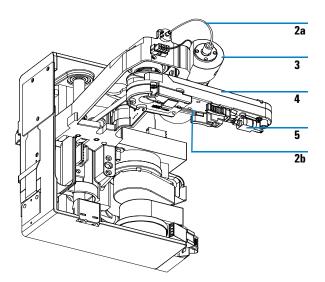


Figure 42 Transport Unit Assembly (Preparative Scale), partly displayed

Transport Unit Assembly (Analytical Scale)

# **Transport Unit Assembly (Analytical Scale)**

ltem	Description	Part Number
1	Transport unit assembly <b>(analytical scale)</b> , includes items 2 and 4	G1364-60019
2a, b	Tubing - diverter valve to needle, 0.25 mm ID (order full kit)	G1364-68712
3	Diverter valve assembly	G1364-61901
4	Needle carrier assembly analytical scale	G1364-60022
5	Needle assembly analytical scale (50mm needle)	G1367-87200
	Needle assembly <b>semi-preparative scale (20mm needle)</b>	G1364-87202
3	Wellplate adapter	G1364-60021

### Table 29 Transport Unit Assembly (Analytical Scale)

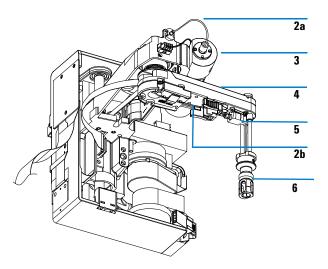


Figure 43 Transport Unit Assembly (Analytical Scale), partly displayed

5 Parts and Materials Needle Assemblies

# **Needle Assemblies**

ltem	Description	Part Number
1	Needle assembly preparative scale	G1364-87201
2	Needle assembly <b>semi preparative scale (20 mm needle)</b> For use in the analytical scale fraction collector at flow rates > 10 ml/min. or with high test tubes (48 mm - 75mm).	G1364-87202
3	Needle assembly <b>analytical scale (50 mm needle)</b>	G1367-87200
	Delay Calibration Adapter (not shown)	G1364-87301

Table 30 Needle Assemblie	s
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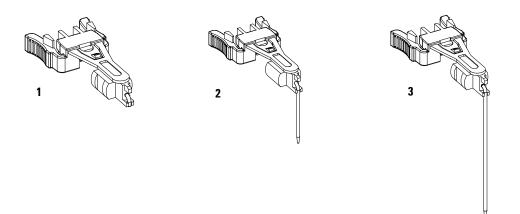


Figure 44 Needle Assemblies

## **Diverter-Valve Assembly**

ltem	Description	Part Number
1	Diverter-valve assembly	G1364-61901
2	PIN screw	0515-1211

**Table 31** Diverter-Valve Assembly (preparative and analytical scale)

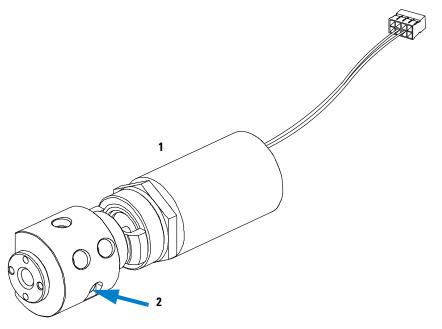


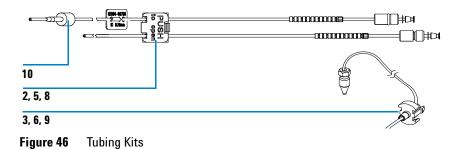
Figure 45 Diverter-Valve Assembly

### 5 Parts and Materials Tubing Kits

# **Tubing Kits**

ltem	Description	Part Number
1	Tubing kit <b>preparative scale 0.8 mm ID</b> (consists of items 2,and 3)	G1364-68711
2	Inlet/waste tubing assembly preparative scale 0.8 mm ID	Order Item 1
3	Diverter valve to needle tubing preparative scale 0.8 mm ID	Order Item 1
4	Tubing kit <b>analytical scale 0.25 mm ID</b> (consists of items 5,and 6)	G1364-68712
5	Inlet/waste tubing assembly <b>analytical scale 0.25 mm ID</b>	Order Item 4
6	Injection valve to needle tubing analytical scale 0.25 mm ID	Order Item 4
7	Low dispersion tubing kit <b>analytical scale 0.15 mm ID</b> for flow rates < 1 ml/min	G1364-68723
8	Inlet/waste tubing assembly <b>analytical scale 0.15 mm ID</b>	Order Item 7
9	injection valve to needle tubing analytical scale 0.15 mm ID	Order Item 7
10	Finger tight fitting (pack of 2)	0100-1516

### Table 32 Tubing Kits



# **Internal Tray Assembly**

ltem	Description	Part Number
1	Internal tray assembly <b>analytical scale</b> , includes items 2 – 7	G1364-63124
	Internal tray assembly <b>preparative scale</b> , includes items 2 – 7	G1364-63123
2	Screw-seat-adapter	5022-2200
3	Seal funnel (reorder No., pack. of 10)	G1364-68730
4a	Waste Tubing Kit 0.5T (analytical scale)	G1364-86708
4b	Waste Tubing Kit 0.8T (preparative scale)	G1364-86719
5	Funnel coupler	G1364-43201
6	Tray internal	Order Item 1
7	Flow delay sensor (not shown)	Order Item 1

 Table 33
 Internal Tray Assembly (Analytical and Preparative Scale)

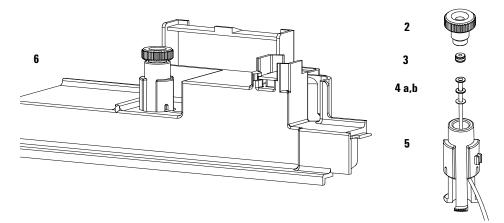


Figure 47 Internal Tray Assembly (Analytical Scale)

**Fraction Collector Accessory Kit** 

## **Fraction Collector Accessory Kit**

Description	Quantity	Part Number
Wrench, open end, 4mm	1	8710-1534
Wrench, open end, 1/4-5/16 inch	1	8710-0510
Hex key 2.0 mm	1	8710-2476
Finger tight fittings, 1/16" f-120 <sup>*</sup>	3	0100-1516
Waste tubing (1.2 m) <sup>†</sup>	1	5062-2463
Y-Type Fitting for waste tubing <sup>‡</sup>	1	5065-9971
CAN cable, 1 m	1	5181-1519
Air channel adapter	1	G1329-43200
Sticking clamp for corrugated waste tubing (large)**	3	5065-9976
Sticking clamp for waste tubing (small) <sup>††</sup>	3	5065-9976
Tray for 15 x 6 ml vials	1	G1313-44513
Tray for 40 x 2 ml vials	1	G1313-44512

### Table 34 Fraction Collector Accessory Kit Contents G1364-68725

\* Reorder gives pack of 2

† Reorder gives 5 m

‡ Reorder gives pack of 10

\*\* Reorder gives 10 clamps for corrugated waste tubing (large))

tt Reorder gives 10 clamps for waste tubing (small)



Agilent 1260 Infinity Fraction Collectors User Manual

# 6 Specifications

Performance Specifications for the Fraction Collectors 142



Agilent Technologies

# **Performance Specifications for the Fraction Collectors**

Туре	Specification
Trigger modes	Time slices, Peak (threshold, up- / downslope), Timetable (combination of time intervals and peak) and Manual trigger (supported only with Agilent Instant Pilot G4208A) Agilent 1260 Infinity DAD/MWD detectors (G1315C,D and G4212B/G1365C,D), the Agilent 1260 Infinity Fluorescence Detector (G1321A) and the Agilent G1946C/D, G1956A/B LC-MSD are fully supported other detectors can be used but are not supported for fraction collection.
Operating modes	Discrete fractions: default mode for all vessels. The flow is diverted to waste, while moving from one vessel position to the next vessel position Continuous flow: optional, available only when using well plates. It is possible to move from one well plate position to the next one without diverting the flow into the well plate to waste
Fraction capacities and trays	4 x well-plates full tray (MTP) <sup>*</sup> (for use with deep well plates, only) 2 × well-plates std. tray (MTP) (for use with deep well plates, only) + 10 × 2 ml vials <sup>*</sup> (+ 1 half tray) 100 x 2 ml in std. tray (+ 1 half tray) <sup>*</sup> 3 x 40 x 2 ml in half tray <sup>*</sup> 3 x 15 x 6 ml in half tray <sup>*</sup> Full tray with 40 test tubes (30 mm OD, max. height 100 mm, ~45 ml / tube) Full tray with 60 test tubes (25 mm OD, max. height 100 mm, ~25 ml / tube) Full tray with 126 test tubes (16 mm OD, max. height 100 mm, ~12 ml / tube) Full tray with 215 test tubes (12 mm OD, max. height 100 mm, ~7 ml / tube) Installed trays are automatically detected and identified. For the with uncapped vials, tests tubes and well plates, only!
Test tube / plate sizes	Minimum 48 mm to 100 mm maximum
Maximum tube volume	ca. 45 ml

### Table 35 performance Specifications Agilent 1260 Infinity Preparative-scale Fraction Collector (G1364B)

6

**Performance Specifications for the Fraction Collectors** 

Туре	Specification
Maximum flow rate	100 ml / min (depending on viscosity and generated back pressure, max. 6 bar at the diverter valve)
Delay volumes [µl]	Fraction collector inlet to diverter valve: ~500 (typical, depends on length of the tubing) Diverter valve: ~15 Diverter valve to needle: ~110 Needle: ~5
Delay calibration sensor	Single wavelength absorbance detector working at 654 nm, consisting of a LED and a photo diode
Diverter valve	3/2 Diverter valve with low internal volume (15 $\mu$ l), switching time < 100 ms, maximum operating pressure 6 bar
cooling	Optional (with additional G1330B), performance depending on ambient conditions and the volume of collected fractions
maximum capacity	3 fraction collectors in parallel plus one recovery fraction collector connected via 12-Position, 13-Port Selector valve (PN G1160A)
GLP features	Early maintenance feedback (EMF), electronic records of maintenance and errors
Interfaces	Controller-area network (CAN). optional; LAN or external contacts interface RS232C, APG-remote (for remote start / stop signals to / from other modules) Interface to G1330B Thermostat CAN-DC-out for operation of Agilent approved external devices like valves
Safety features	Leak detection and safe leak handling, error detection and display, exhaust fan for fume extraction of hazardous vapors

Table 35	performance Specifications Agilent 1260 Infinity Preparative-scale Fraction
	Collector (G1364B) (continued)

\* Vials can be used as recommended by Agilent Technologies (see "List of Recommended Vials and Caps" on page 129 and "List of Recommended Plates and Closing Mats" on page 132) but must be uncapped. Only the 96 deep well-plates can be used (without closing mats, see "List of Recommended Plates and Closing Mats" on page 132)

### NOTE

Only one type of well-plates can be used at a time in one tray.

### 6 Specifications

**Performance Specifications for the Fraction Collectors** 

Туре	Specification
Trigger modes	Time slices, Peak (threshold, up- / downslope), Timetable (combination of time intervals and peak) and Manual trigger (supported only with Agilent Instant Pilot G4208A) Agilent 1260 Infinity DAD/MWD detectors (G1315C,D; G4212B/ G1365 C,D), the Agilent 1260 Infinity Fluorescence Detector (G1321A) and the Agilent G1946C/D, G1956A/B LC-MSD are fully supported other detectors can be used but are not supported for fraction collection.
Operating modes	Discrete fractions: default mode for all vessels. The flow is diverted to waste, while moving from one vessel position to the next vessel position Continuous flow: optional, available only when using the deep well plates. It is possible to move from one well plate position to the next one without diverting the flow into the well plate to waste Needle into location: Needle pushes into the vessel as deep as specified, for the use with capped vials and test tubes and well plates with closing mats Droplet setup mode: The tip of the fraction collector needle will initially move down to the bottom of the well. Then it will slowly move upwards while the fraction is collected. The droplet setup mode enables the fraction collector to collect small fractions without bubbles.

### Table 36 Performance Specifications Agilent 1260 Infinity Analytical-scale Fraction Collector (G1364C)

**Performance Specifications for the Fraction Collectors** 

Туре	Specification	
Fraction vessel capacities and trays	<ul> <li>4 x well-plates full tray (MTP)<sup>*</sup></li> <li>2 x well-plates std. tray + 10 funnels with external containers<sup>*</sup> (+ 1 half tray)</li> <li>2 × well-plates std. tray (MTP) + 10 × 2 ml vials<sup>*</sup> (+ 1 half tray)</li> <li>100 x 2 ml in std. tray (+ 1 half tray)<sup>*</sup></li> <li>3 x 40 x 2 ml in half tray<sup>*</sup></li> <li>3 x 40 funnels in half tray<sup>*</sup></li> <li>3 x 40 funnels in half tray<sup>*</sup></li> <li>3 x 15 x 6 ml in half tray<sup>*</sup></li> <li>Full tray with 40 test tubes (30 mm OD, max. height 48 mm, ~20 ml vol.)</li> <li>Full tray with 60 test tubes (25 mm OD, max. height 48 mm)</li> <li>Full tray with 126 test tubes (16 mm OD, max. height 48 mm)</li> <li>Full tray with 215 test tubes (12 mm OD, max. height 48 mm)</li> <li>Installed trays are automatically detected and identified. Installed plates and vials can be detected when operating in the needle into location mode</li> <li>* max. height can be extended by using the short needle assembly G1364-87202</li> </ul>	
Maximum tube / plate height	48 mm with long needle assembly G1367-87200 75 mm with short needle assembly G1364-87202	
Maximum tube volume	ca. 20 ml with 48 mm test tubes, ca. 30 ml with 75 mm test tubes or unlimited, if funnels are used with external containers.	
Maximum flow rate	10 ml / min (depending on viscosity and generated back pressure, max. 6 bar at the diverter valve). The analytical scale fraction collector can be modified for flow rates > 10 ml/min.	
Delay volumes [µl]	Fraction collector inlet to diverter valve: ~50 (typical, depends on the length of the tubing) Diverter valve: ~15 Diverter valve to needle: ~10 Needle: ~4	
Delay calibration sensor	Single wavelength absorbance detector working at 654 nm, consisting of a LED and a photo diode	
Diverter valve	$3/2$ Diverter valve with low internal volume (15 $\mu$ l), switching time $<$ 100 ms, maximum operating pressure 6 bar	

### Table 36 Performance Specifications Agilent 1260 Infinity Analytical-scale Fraction Collector (G1364C) (continued)

### 6 Specifications

**Performance Specifications for the Fraction Collectors** 

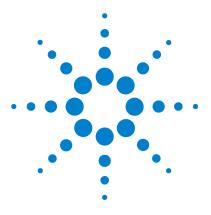
Туре	Specification	
Cooling	Optional (with additional G1330B), performance depending on ambient conditions and the volume of collected fractions	
Maximum capacity	3 fraction collectors in parallel plus one recovery fraction collector connected via 12-Position, 13-Port Selector valve (PN G1160A)	
GLP features	Early maintenance feedback (EMF), electronic records of maintenance and errors	
Interfaces	<ul> <li>Controller-area network (CAN).</li> <li>optional; LAN or external contacts interface</li> <li>RS232C,</li> <li>APG-remote (for remote start / stop signals to / from other modules)</li> <li>Interface to G1330B Thermostat</li> <li>CAN-DC-out for operation of Agilent approved external devices like valves</li> </ul>	
Safety features	Leak detection and safe leak handling, error detection and display, exhaust fan for fume extraction of hazardous vapors	

 
 Table 36
 Performance Specifications Agilent 1260 Infinity Analytical-scale Fraction Collector (G1364C) (continued)

 Vials and well-plates and capped vials and well plates with closing mats can be used as recommended by Agilent Technologies (see "List of Recommended Vials and Caps" on page 129 and "List of Recommended Plates and Closing Mats" on page 132)

### NOTE

Only one type of well-plates can be used at a time in one tray.



Agilent 1260 Infinity Fraction Collectors User Manual

## A Safety Information

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Agilent Technologies

A Safety Information Safety Information

## **Safety Information**

The following general safety precautions must be observed during all phases of operation, service, and repair of this instrument. Failure to comply with these precautions or with specific warnings elsewhere in this manual violates safety standards of design, manufacture, and intended use of the instrument. Agilent Technologies assumes no liability for the customer's failure to comply with these requirements.

### General

This is a Safety Class I instrument (provided with terminal for protective earthing) and has been manufactured and tested according to international safety standards.

#### WARNING

If you are using flammable solvents, remove the well-plates from the tray when you turn off the sampler. You avoid the risk of building explosive gas mixtures in the tray compartment.

**WARNING** If you are using flammable solvents, cover the well-plates with closing mats to avoid the risk of building explosive gas mixtures.

### WARNING

After a leak in the sampler, make sure the leak plane is cleaned and dry.

### Operation

Before applying power, comply with the installation section. Additionally the following must be observed.

Do not remove instrument covers when operating. Before the instrument is switched on, all protective earth terminals, extension cords, auto-transformers, and devices connected to it must be connected to a protective earth via a ground socket. Any interruption of the protective earth grounding will cause a potential shock hazard that could result in serious personal injury. Whenever it is likely that the protection has been impaired, the instrument must be made inoperative and be secured against any intended operation.

Make sure that only fuses with the required rated current and of the specified type (normal blow, time delay, and so on) are used for replacement. The use of repaired fuses and the short-circuiting of fuseholders must be avoided.

# **WARNING** Any adjustment, maintenance, and repair of the opened instrument under voltage is forbidden.

### WARNING

# Disconnect the instrument from the line and unplug the power cord before maintenance.

Do not operate the instrument in the presence of flammable gases or fumes. Operation of any electrical instrument in such an environment constitutes a definite safety hazard.

Do not install substitute parts or make any unauthorized modification to the instrument.

Capacitors inside the instrument may still be charged, even though the instrument has been disconnected from its source of supply. Dangerous voltages, capable of causing serious personal injury, are present in this instrument. Use extreme caution when handling, testing and adjusting.

## **Safety Symbols**

Table 37 shows safety symbols used on the instrument and in the manuals.

Table 37	Safety S	Symbols
----------	----------	---------

Symbol	Description	
	The apparatus is marked with this symbol when the user should refer to the instruction manual in order to prevent risk of harm to the operator and to protect the apparatus against damage.	
4	Indicates dangerous voltages.	
	Indicates a protected conductor terminal.	
	Eye damage may result from directly viewing the light produced by the Xenon flash lamp used in this product. Always turn the xenon flash lamp off before removing it.	

### WARNING

A warning alerts you to situations that could cause physical injury or damage to the equipment. Do not proceed beyond a warning until you have fully understood and met the indicated conditions.

### CAUTION

A caution alerts you to situations that could cause a possible loss of data. Do not proceed beyond a caution until you have fully understood and met the indicated conditions.

## **Lithium Batteries Information**

### WARNING

Danger of explosion if battery is incorrectly replaced. Replace only with the same or equivalent type recommended by the equipment manufacturer. Lithium batteries may not be disposed-off into the domestic waste.

Transportation of discharged Lithium batteries through carriers regulated by IATA/ICAO, ADR, RID, IMDG is not allowed. Discharged Lithium batteries shall be disposed off locally according to national waste disposal regulations for batteries.

### WARNING

Lithiumbatteri - Eksplosionsfare ved fejlagtic handtering. Udskiftning ma kun ske med batteri af samme fabrikat og type. Lever det brugte batteri tilbage til leverandoren.

### WARNING

Lithiumbatteri - Eksplosionsfare. Ved udskiftning benyttes kun batteri som anbefalt av apparatfabrikanten. Brukt batteri returneres appararleverandoren.

### NOTE

Bij dit apparaat zijn batterijen geleverd. Wanneer deze leeg zijn, moet u ze niet weggooien maar inleveren als KCA.





## **Radio Interference**

Never use cables other than the ones supplied by Agilent Technologies to ensure proper functionality and compliance with safety or EMC regulations.

### **Test and Measurement**

If test and measurement equipment is operated with equipment unscreened cables and/or used for measurements on open set-ups, the user has to assure that under operating conditions the radio interference limits are still met within the premises.

## **Sound Emission**

## **Manufacturer's Declaration**

This statement is provided to comply with the requirements of the German Sound Emission Directive of 18 January 1991.

This product has a sound pressure emission (at the operator position) < 70 dB.

- Sound Pressure Lp < 70 dB (A)
- At Operator Position
- Normal Operation
- According to ISO 7779:1988/EN 27779/1991 (Type Test)

## **Solvent Information**

Observe the following recommendations on the use of solvents.

### WARNING

This instrument should only be used with solvents that have an ignition temperature higher than 200°C!

### **Solvents**

Brown glass ware can avoid growth of algae.

Always filter solvents, small particles can permanently block the capillaries. Avoid the use of the following steel-corrosive solvents:

- Solutions of alkali halides and their respective acids (for example, lithium iodide, potassium chloride, and so on).
- High concentrations of inorganic acids like nitric acid, sulfuric acid especially at higher temperatures (replace, if your chromatography method allows, by phosphoric acid or phosphate buffer which are less corrosive against stainless steel).
- Halogenated solvents or mixtures which form radicals and/or acids, for example:

 $2CHCl_3 + O_2 \rightarrow 2COCl_2 + 2HCl$ 

This reaction, in which stainless steel probably acts as a catalyst, occurs quickly with dried chloroform if the drying process removes the stabilizing alcohol.

- Chromatographic grade ethers, which can contain peroxides (for example, THF, dioxane, di-isopropylether) such ethers should be filtered through dry aluminium oxide which adsorbs the peroxides.
- Solutions of organic acids (acetic acid, formic acid, and so on) in organic solvents. For example, a 1-% solution of acetic acid in methanol will attack steel.

- Solutions containing strong complexing agents (for example, EDTA, ethylene diamine tetra-acetic acid).
- Mixtures of carbon tetrachloride with 2-propanol or THF.

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For the latest information on products and services visit our worldwide web site on the Internet at:

http://www.agilent.com

Select "Products" - "Chemical Analysis"

It will provide also the latest firmware of the Agilent 1260 Infinity modules for download.

### A Safety Information

**Agilent Technologies on Internet** 

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### In This Book

This manual contains technical reference information about the Agilent 1260 Infinity Fraction Collectors. The manual describes the following:

- configuration and operation of the fraction collector,
- troubleshooting and test functions,
- simple repairs
- parts and materials,
- specifications,
- safety information.

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