

# Agilent 1260 Infinity Bio-inert High Performance Autosampler



# User Manual

# Notices

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## In This Guide

This manual covers the Agilent 1260 Infinity Bio-inert High Performance Autosampler (G5667A)

### **1** Introduction

This chapter gives an introduction to the autosampler.

### 2 Site Requirements and Specifications

This chapter provides information on environmental requirements, physical and performance specifications.

#### **3** Installing the Autosampler

This chapter provides information on unpacking, checking on completeness, stack considerations and installation of the autosampler.

#### 4 LAN Configuration

This chapter provides information on connecting the autosampler to the Agilent ChemStation PC.

#### 5 Using the Module

This chapter provides information on how to set up the autosampler for an analysis and explains the basic settings.

#### 6 Optimizing Performance

This chapter gives hints on how to optimize the performance or use additional devices.

### 7 Troubleshooting and Diagnostics

This chapter gives an overview about the troubleshooting and diagnostic features and the different user interfaces.

### 8 Error Information

This chapter describes the meaning of error messages, and provides information on probable causes and suggested actions how to recover from error conditions.

### 9 Test Functions

This chapter describes the tests for the module.

### **10 Maintenance**

This chapter describes the maintenance of the Autosampler

### **11 Parts for Maintenance**

This chapter provides information on parts material required for the module.

### **12 Identifying Cables**

This chapter provides information on cables used with the 1260 series of HPLC modules.

### **13 Hardware Information**

This chapter describes the autosampler in more detail on hardware and electronics.

### **14 Appendix**

This chapter provides addition information on safety, legal and web.

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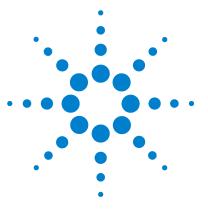
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# Introduction

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This chapter gives an introduction to the autosampler.



1 Introduction Features

# **Features**

The Agilent 1260 Infinity Bio-inert High Performance Autosampler features an increased pressure range (up to 600 bar) enabling the use of today's column technology (sub-two-micron narrow bore columns) with the Agilent 1260 Infinity LC System. Increased robustness is achieved by optimized new parts, high speed with lowest carry-over by flow through design, increased sample injection speed for high sample throughput, increased productivity by using overlapped injection mode and flexible and convenient sample handling with different types of sample containers, such as vials and well plates. Using 384-well plates allows you to process up to 768 samples unattended.

For specifications, see "Specifications" on page 28

**NOTE** This 1260 Infinity Autosampler has been introduced together with the Agilent 1260 Infinity Liquid Chromatograph.

# **Bio-inert Materials**

For the Agilent 1260 Infinity Bio-inert LC system, Agilent Technologies uses highest quality materials in the flow path (also referred to as wetted parts), which are widely accepted by life scientists, as they are known for optimum inertness to biological samples and ensure best compatibility with common samples and solvents over a wide pH range. Explicitly, the complete flow path is free of stainless steel and free of other alloys containing metals such as iron, nickel, cobalt, chromium, molybdenum or copper, which can interfere with biological samples. The flow downstream of the sample introduction contains no metals whatsoever.

Module	Materials
Agilent 1260 Infinity Bio-inert Quaternary Pump (G5611A)	Titanium, gold, platinum-iridium, ceramic, ruby, PTFE, PEEK
Agilent 1260 Infinity Bio-inert High-Performance Autosampler (G5667A)	Upstream of sample introduction: • Titanium, gold, PTFE, PEEK, ceramic
	Downstream of sample introduction: • PEEK, ceramic
Agilent 1260 Infinity Bio-inert Manual Injector (G5628A)	PEEK, ceramic
Agilent 1260 Infinity Bio-inert Analytical Fraction Collector (G5664A)	PEEK, ceramic, PTFE

Table 1	Bio-inert materials used in Agilent 1260 Infinity Systems

### 1 Introduction

**Bio-inert Materials** 

Module	Materials
Bio-inert Flow Cells:	
Standard flow cell bio-inert, 10mm, 13 µl, 120 bar (12 MPa) for MWD/DAD, includes Capillary Kit Flow Cells BIO (p/n G5615-68755) (p/n G5615-60022) ( <i>for Agilent 1260 Infinity Diode Array Detectors DAD G1315C/D</i> )	PEEK, ceramic, sapphire, PTFE
Max-Light Cartridge Cell Bio-inert (10 mm, V(σ) 1.0 μl) (p/n G5615-60018) and	PEEK, fused silica
Max-Light Cartridge Cell Bio-inert (60 mm, V(σ) 4.0 μl) (p/n G5615-60017) (for Agilent 1200 Infinity Series Diode Array Detectors DAD G4212A/B)	
Bio-inert flow cell, 8 μL, 20 bar (pH 1–12) includes Capillary Kit Flow Cells BIO (p/n G5615-68755)) (p/n G5615-60005) (for Agilent 1260 Infinity Fluorescence Detector FLD G1321B)	PEEK, fused silica, PTFE
Bio-inert heat-exchanger G5616-60050 (for Agilent 1290 Infinity Thermostatted Column Compartment G1316C)	PEEK (steel-cladded)
Bio-inert Valve heads	G4235A, G5631A, G5639A: PEEK, ceramic (Al <sub>2</sub> O <sub>3</sub> based)
Bio-inert Connection capillaries	Upstream of sample introduction: • Titanium
	<ul> <li>Downstream of sample introduction:</li> <li>Agilent uses stainless-steel-cladded PEEK capillaries, which keep the flow path free of steel and provide pressure stability to more than</li> </ul>

### Table 1 Bio-inert materials used in Agilent 1260 Infinity Systems

### NOTE

To ensure optimum bio-compatibility of your Agilent 1260 Infinity Bio-inert LC system, do not include non-inert standard modules or parts to the flow path. Do not use any parts that are not labeled as Agilent "Bio-inert". For solvent compatibility of these materials, see "Solvent information for parts of the 1260 Infinity Bio-inert LC system" on page 49.

# **Overview of the Module**

The Autosampler transport mechanism uses an X-Z-theta robot to optimize the positioning of the sampling arm on the well plate. Once the sampling arm is positioned over the programmed sample position, the programmed sample volume is drawn by the metering device into the sampling needle. The sampling arm then moves to the injection position where the sample is flushed onto the column.

The Autosampler employ a vial/plate pusher mechanism to hold down the vial or the plate while the needle is drawn back from the sample vessel (a must in the case a septum is used). This vial/plate pusher employs a sensor to detect the presence of a plate and to ensure accurate movement regardless of plate used.

All axes of the transport mechanism (x-,z-,theta-robot) are driven by stepper-motors. Optical encoders ensure the correct operation of the movement.

The standard metering device provides injection volumes from  $0.1 - 100 \mu$ L. The entire flow path including the metering device is always flushed by the mobile phase after injection for minimum internal carry-over.

An additional needle flush station with a peristaltic pump is installed to wash the outside of the needle. This reduces the already low carry-over for very sensitive analysis.

The bottle containing the mobile phase for the wash procedure will be located in the solvent bottle cabinet. Produced waste during this operation is channeled safely away through a waste drain.

The six-port (only 5 ports are used) injection valve unit is driven by a high-speed hybrid stepper motor. During the sampling sequence, the valve unit bypasses the autosampler, and connects flow from the pump to the column directly. During injection and analysis, the valve unit directs the flow through the autosampler which ensures that the entire sample is injected onto the column, and that the metering unit and needle are always free from sample residue before the next sampling sequence begins.

Control of the vial/plate temperature in the thermostatted autosampler is achieved using an additional Agilent 1290 Infinity Series module; the Agilent

1290 Infinity Series thermostat for ALS/FC/Spotter. The thermostat contains Peltier-controlled heat-exchangers. A fan draws air from the area above the sample vial tray of the autosampler. It is then blown through the fins of the cooling/heating module. There it is cooled or heated according the temperature setting. The thermostatted air enters the autosampler through a recess underneath the special designed sample tray. The air is then distributed evenly through the sample tray ensuring effective temperature control, regardless of how many vials are in the tray. In cooling mode condensation is generated on the cooled side of the Peltier elements. This condensed water is safely guided into a waste bottle for condensed water.

# Autosampler Principle

The movements of the autosampler components during the sampling sequence are monitored continuously by the autosampler processor. The processor defines specific time windows and mechanical ranges for each movement. If a specific step of the sampling sequence is not completed successfully, an error message is generated. Solvent is bypassed from the autosampler by the injection valve during the sampling sequence. The needle moves to the desired sample position and is lowered into the sample liquid in the sample to allow the metering device to draw up the desired volume by moving its plunger back a certain distance. The needle is then raised again and moved onto the seat to close the sample loop. Sample is applied to the column when the injection valve returns to the mainpass position at the end of the sampling sequence.

The standard sampling sequence occurs in the following order:

- **1** The injection valve switches to the bypass position.
- **2** The plunger of the metering device moves to the initialization position.
- **3** The needle lock moves up.
- **4** The needle moves to the desired sample vial (or well plate) position.
- **5** The needle lowers into the sample vial (or well plate).
- **6** The metering device draws the preset sample volume.
- 7 The needle lifts out of the sample vial (or well plate).
- 8 The needle is then moved onto the seat to close the sample loop.
- **9** The needle lock moves down.
- **10** The injection cycle is completed when the injection valve switches to the mainpass position.
- If needle wash is required it will be done between step 7 and 8.

### **Injection Sequence**

Before the start of the injection sequence, and during an analysis, the injection valve is in the mainpass position. In this position, the mobile phase flows through the autosampler metering device, sample loop, and needle, ensuring all parts in contact with sample are flushed during the run, thus minimizing carry-over.

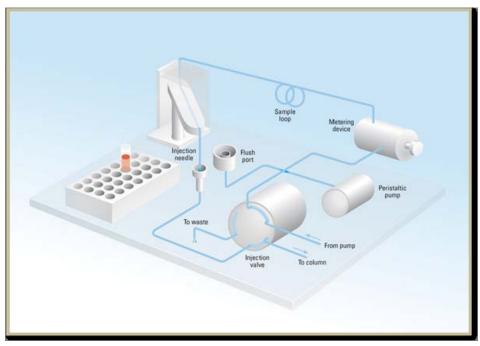


Figure 1 Mainpass Position

When the sample sequence begins, the valve unit switches to the bypass position. Solvent from the pump enters the valve unit at port 1, and flows directly to the column through port 6.

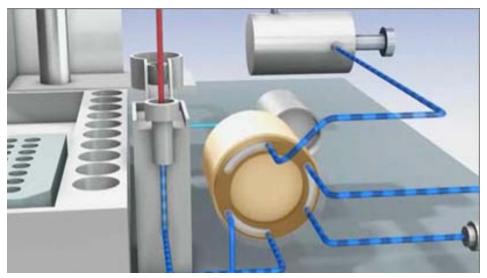
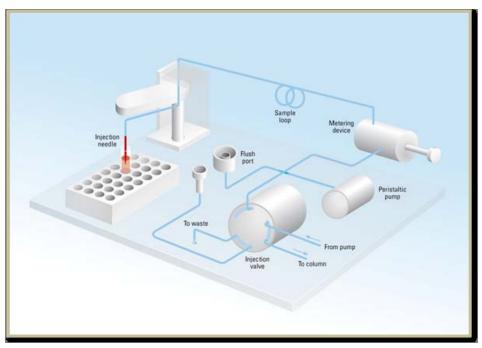


Figure 2 Bypass Position

The standard injection starts with *draw sample from vial*. In order to do this the needle moves to the desired sample position and is lowered into the sample liquid in the sample to allow the metering device to draw up the desired volume by moving its plunger back a certain distance. The needle is then raised again and moved onto the seat to close the sample loop. In case of an injector program several steps are interspersed at this point.

**Autosampler Principle** 



**Figure 3** Drawing the Sample

**Flush the Needle** Before injection and to reduce the carry-over for very sensitive analysis, the outside of the needle can be washed in a flush port located behind the injector port on the sampling unit. As soon as the needle is on the flush port a peristaltic pump delivers some solvent during a defined time to clean the outside of the needle. At the end of this process the needle returns to the injection port.

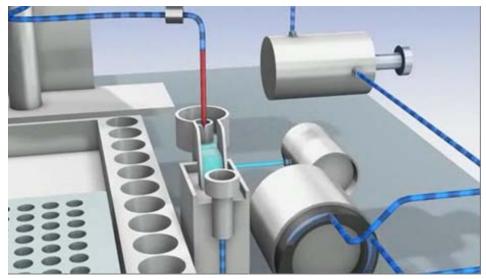


Figure 4 Flush the needle

**Inject-and-Run** The final step is the inject-and-run step. The six-port valve is switched to the main-pass position, and directs the flow back through the sample loop, which now contains a certain amount of sample. The solvent flow transports the sample onto the column, and separation begins. This is the beginning of a *run* within an analysis. In this stage, all major performance-influencing hardware is flushed internally by the solvent flow. For standard applications no additional flushing procedure is required.

### 1 Introduction

**Autosampler Principle** 

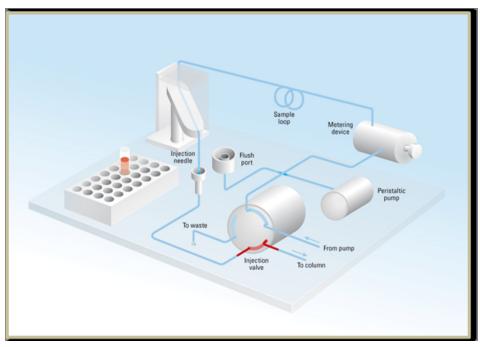


Figure 5 Inject and Run

1

# **Early Maintenance Feedback**

Maintenance requires the exchange of components which are subject to wear or stress. Ideally, the frequency at which components are exchanged should be based on the intensity of usage of the module and the analytical conditions, and not on a predefined time interval. The early maintenance feedback (**EMF**) feature monitors the usage of specific components in the instrument, and provides feedback when the user-selectable limits have been exceeded. The visual feedback in the user interface provides an indication that maintenance procedures should be scheduled.

### **EMF Counters**

**EMF counters** increment with use and can be assigned a maximum limit which provides visual feedback in the user interface when the limit is exceeded. Some counters can be reset to zero after the required maintenance procedure.

### Using the EMF Counters

The user-settable **EMF** limits for the **EMF Counters** enable the early maintenance feedback to be adapted to specific user requirements. The useful maintenance cycle is dependent on the requirements for use. Therefore, the definition of the maximum limits need to be determined based on the specific operating conditions of the instrument.

### Setting the EMF Limits

The setting of the **EMF** limits must be optimized over one or two maintenance cycles. Initially the default **EMF** limits should be set. When instrument performance indicates maintenance is necessary, take note of the values displayed by the **EMF counters**. Enter these values (or values slightly less than the displayed values) as **EMF** limits, and then reset the **EMF counters** to zero. The next time the **EMF counters** exceed the new **EMF** limits, the **EMF** flag will be displayed, providing a reminder that maintenance needs to be scheduled.

# **Instrument Layout**

The industrial design of the module incorporates several innovative features. It uses Agilent's E-PAC concept for the packaging of electronics and mechanical assemblies. This concept is based upon the use of expanded polypropylene (EPP) layers of foam plastic spacers in which the mechanical and electronic boards components of the module are placed. This pack is then housed in a metal inner cabinet which is enclosed by a plastic external cabinet. The advantages of this packaging technology are:

- virtual elimination of fixing screws, bolts or ties, reducing the number of components and increasing the speed of assembly/disassembly,
- the plastic layers have air channels molded into them so that cooling air can be guided exactly to the required locations,
- the plastic layers help cushion the electronic and mechanical parts from physical shock, and
- the metal inner cabinet shields the internal electronics from electromagnetic interference and also helps to reduce or eliminate radio frequency emissions from the instrument itself.



# **Site Requirements and Specifications**

Site Requirements24Physical Specifications27Specifications28

This chapter provides information on environmental requirements, physical and performance specifications.



2 Site Requirements and Specifications Site Requirements

## **Site Requirements**

A suitable environment is important to ensure optimal performance of the module.

### **Power Consideration**

The module power supply has wide ranging capabilities and accepts any line voltage in the range mentioned in Table 2 on page 27. Consequently, there is no voltage selector in the rear of the module. There are also no externally accessible fuses, because automatic electronic fuses are implemented in the power supply.

### WARNING

Module is partially energized when switched off, as long as the power cord is plugged in.

Repair work at the module can lead to personal injuries, e.g. shock hazard, when the cover is opened and the module is connected to power.

- → Make sure that it is always possible to access the power plug.
- → Remove the power cable from the instrument before opening the cover.
- → Do not connect the power cable to the Instrument while the covers are removed.

### WARNING

### Incorrect line voltage at the module

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

→ Connect your module to the specified line voltage.

### CAUTION

Inaccessible power plug.

In case of emergency it must be possible to disconnect the instrument from the power line at any time.

- → Make sure the power connector of the instrument can be easily reached and unplugged.
- Provide sufficient space behind the power socket of the instrument to unplug the cable.

### **Power Cords**

Different power cords are offered as options with the module. The female end of all power cords is identical. It plugs into the power-input socket at the rear. The male end of each power cord is different and designed to match the wall socket of a particular country or region.

### WARNING

### Absence of ground connection or use of unspecified power cord

The absence of ground connection or the use of unspecified power cord can lead to electric shock or short circuit.

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

### WARNING

### Use of unsupplied cables

Using cables not supplied by Agilent Technologies can lead to damage of the electronic components or personal injury.

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

### 2 Site Requirements and Specifications

**Site Requirements** 

### WARNING

### Unintended use of supplied power cords

Using power cords for unintended purposes can lead to personal injury or damage of electronic equipment.

Never use the power cords that Agilent Technologies supplies with this instrument for any other equipment.

### **Bench Space**

The module dimensions and weight (see Table 2 on page 27) allow you to place the module on almost any desk or laboratory bench. It needs an additional 2.5 cm (1.0 inches) of space on either side and approximately 8 cm (3.1 inches) in the rear for air circulation and electric connections.

If the bench shall carry a complete HPLC system, make sure that the bench is designed to bear the weight of all modules.

The module should be operated in a horizontal position.

### Condensation

### CAUTION

Condensation within the module

Condensation will damage the system electronics.

- → Do not store, ship or use your module under conditions where temperature fluctuations could cause condensation within the module.
- → If your module was shipped in cold weather, leave it in its box and allow it to warm slowly to room temperature to avoid condensation.

# **Physical Specifications**

Туре	Specification	Comments
Weight	15.5 kg (35 lbs)	
Dimensions (height × width × depth)	200 x 345 x 440 mm (8 x 13.5 x 17 inches)	
Line voltage	100 - 240 VAC, ± 10 %	Wide-ranging capability
Line frequency	50 or 60 Hz, ± 5 %	
Power consumption	200 VA / 200 W / 683 BTU	Maximum
Ambient operating temperature	4–55 °C (41–131 °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 (6562 ft)	
Non-operating altitude	Up to 4600 m (15091 ft)	For storing the module
Safety standards: IEC, CSA, UL	Installation category II, Pollution degree 2	For indoor use only.

### Table 2 Physical Specifications

# **Specifications**

Specification
$0.1-100~\mu L$ in 0.1 $\mu L$ increments. Up to 40 $\mu L$ with reduced injection volume kit (hardware modification required). Up to 1500 $\mu L$ with multiple draw (hardware modification required)
Typically < 0.25 % RSD from 5 – 100 μL. Typically < 0.5 % RSD from 2 – 5 μL. Typically < 0.7 % RSD from 1 – 2 μL volume. Measured with injections of caffeine in water
1 % (10 µL, n=10)
Up to 600 bar (8700 psi)
0.2 – 5 cp
2 x well plates (MTP) + 10 x 2.0 mL vials 108 x 2 mL vials in 2 x 54 vial plate plus 10 additional 2 mL vials 30 x 6 mL vials in 2 x 15 vial plate plus 10 additional 2 mL vials 54 Eppendorf tubes (0.5/1.5/2 mL) in 2 x 27 Eppendorf tube plates
Typically < 17 s using the following standard conditions: Default draw speed: 100 $\mu$ L/min. Default eject speed: 100 $\mu$ L/min. Injection volume: 5 $\mu$ L
Typically <0.004 % For measurement conditions see <sup>1</sup> , <sup>2</sup> , <sup>3</sup>
1 – 13 (short term 14 <sup>4</sup> )
Optional with G1330B 4 – 40 °C
Upstream of sample-introduction: • titanium, gold, PTFE, PEEK, ceramic
Downstream of sample-introduction: • PEEK, ceramic

 
 Table 3
 Specifications - Agilent 1260 Infinity Bio-inert High-Performance Autosampler (G5667A)

Table 3	Specifications - Agilent 1260 Infinity Bio-inert High-Performance Autosampler
	(G5667A)

Туре	Specification
GLP features	Early maintenance feedback (EMF), electronic records of maintenance and errors
Communications	Controller-area network (CAN). RS232C, APG-remote standard, optional four external contact closures and BCD vial number output
Safety features Extensive diagnostics can be done with the help of the Con and Agilent LabAdvisor Diagnostic Software, leak detectio leak handling, low voltages in maintenance areas, error det display	

<sup>1</sup> Chromatographic conditions: Column: Agilent ZORBAX SB-C18, 2.1 x 50 mm1.8 μm (p/n 827700-902); mobile phase: A: 0.1 % TFA in water, B: 0.1 % TFA in acetonitrile; isocratic : %B=35 %; flow rate: 0.5 mL/min; temperature: 30 °C

- <sup>2</sup> UV-detection: Sample : 1200 ng/µl chlorhexidine (dissolved in mobile phase A), 1 µL injected and measured on G4212A DAD (10 mm cell); Wavelength: 257 nm +/- 4 nm; ref. 360 nm +/- 16 nm; slit 4 nm, 10 Hz
- <sup>3</sup> MS-detection: Sample : 50 ng/μl chlorhexidine (dissolved in mobile phase A), 1 μL injected and measured on Agilent 6460 QQQ (in specified conditions); MRM 1: 505.5 ? 170 (CE: 36 V); MRM 2: 505.5 ? 201.2 (CE: 20 V); fragmentor: 150 V, delta EMV(+): 200 V
- <sup>4</sup> For solvent compatibility, refer to section "Solvent information for parts of the 1260 Infinity Bio-inert LC system" in the manual

2 Site Requirements and Specifications Specifications



# Installing the Autosampler

Unpacking the Autosampler 32 Damaged Packaging 32 Delivery Checklist 32 Autosampler Accessory Kit Contents 33 Optimizing the Stack Configuration 34 **One Stack Configuration** 35 **Two Stack Configuration** 37 Installing the Autosampler 39 Flow Connections to the Autosampler 41 Installation of stainless steel cladded PEEK capillaries 42

This chapter provides information on unpacking, checking on completeness, stack considerations and installation of the autosampler.



# **Unpacking the Autosampler**

## **Damaged Packaging**

If the delivery packaging shows signs of external damage, please call your Agilent Technologies sales and service office immediately. Inform your service representative that the instrument may have been damaged during shipment.

### CAUTION

### "Defective on arrival" problems

If there are signs of damage, please do not attempt to install the module. Inspection by Agilent is required to evaluate if the instrument is in good condition or damaged.

- → Notify your Agilent sales and service office about the damage.
- An Agilent service representative will inspect the instrument at your site and initiate appropriate actions.

## **Delivery Checklist**

Ensure all parts and materials have been delivered with the autosampler. 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.

 Table 4
 Autosampler Checklist

Description	Quantity
Autosampler	1
Power cable	1
User manual on Documentation CD (part of the shipment - not module specific)	1
Accessory kit	1

# **Autosampler Accessory Kit Contents**

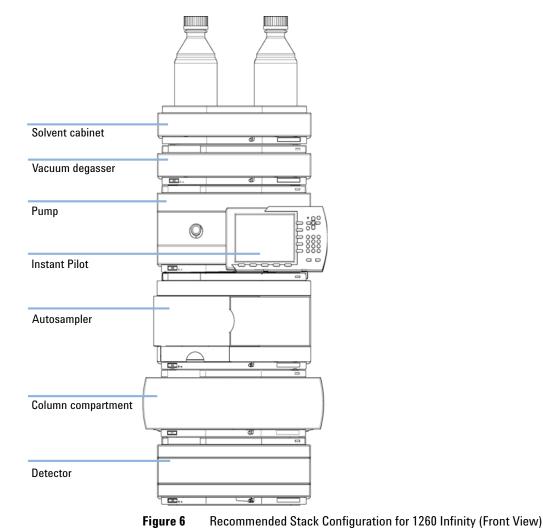
p/n	Description
G5667-68755	Accessory kit (bio-inert)
G5667-60500	PEEK/SST capillary (bio-inert) 0.17 mm ID, 400 mm long
5181-1519	CAN cable, Agilent module to module, 1 m
5063-6527	Tubing assembly, i.d. 6 mm, o.d. 9 mm, 1.2 m (to waste)

# **Optimizing the Stack Configuration**

If your module is part of a complete Agilent 1260 Infinity Liquid Chromatograph, you can ensure optimum performance by installing the following configurations. These configurations optimize the system flow path, ensuring minimum delay volume.

## **One Stack Configuration**

Ensure optimum performance by installing the modules of the Agilent 1260 Infinity LC System in the following configuration (See Figure 6 on page 35 and Figure 7 on page 36). This configuration optimizes the flow path for minimum delay volume and minimizes the bench space required.



### **3** Installing the Autosampler

**Optimizing the Stack Configuration** 

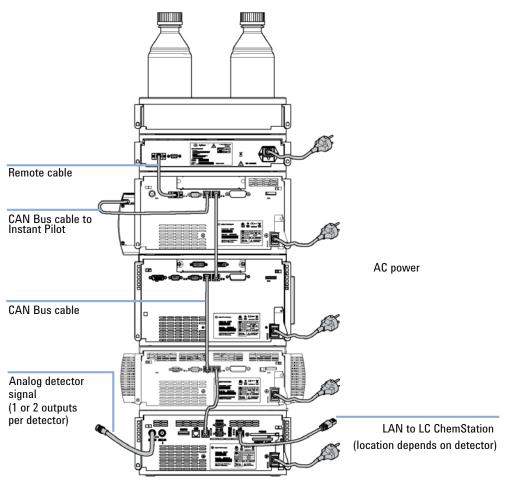


Figure 7 Recommended Stack Configuration for 1260 Infinity (Rear View)

# **Two Stack Configuration**

To avoid excessive height of the stack when the autosampler thermostat is added to the system it is recommended to form two stacks. Some users prefer the lower height of this arrangement even without the autosampler thermostat. A slightly longer capillary is required between the pump and autosampler. (See Figure 8 on page 37 and Figure 9 on page 38).

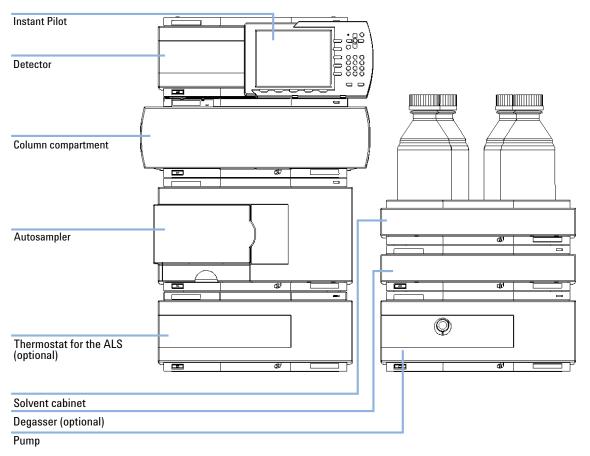
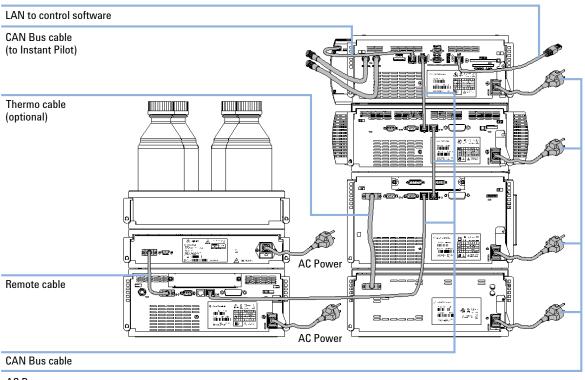


Figure 8 Recommended Two Stack Configuration for 1260 Infinity (Front View)

**Optimizing the Stack Configuration** 



AC Power

Figure 9 Recommended Two Stack Configuration for 1260 Infinity (Rear View)

Parts required	<b>Description</b> Autosampler Power cord
Hardware required	Other cables see below and section "Cable Overview" on page 164
Software required	ChemStation and/or Instant Pilot G4208A with the appropriate revisions.
WARNING	Module is partially energized when switched off, as long as the power cord is plugged in.
	Repair work at the module can lead to personal injuries, e.g. shock hazard, when the cover is opened and the module is connected to power.
	→ Make sure that it is always possible to access the power plug.
	ightarrow Remove the power cable from the instrument before opening the cover.
	→ Do not connect the power cable to the Instrument while the covers are removed.
CAUTION	"Defective on arrival" problems
	If there are signs of damage, please do not attempt to install the module. Inspection by Agilent is required to evaluate if the instrument is in good condition or damaged.
	→ Notify your Agilent sales and service office about the damage.
	→ An Agilent service representative will inspect the instrument at your site and initiate appropriate actions.
	1 Place the Autosampler in the stack, see "Optimizing the Stack Configuration" on page 34.
	<b>2</b> Ensure the power switch on the front of the module is OFF (switch stands out).

Image: Section of the section of t	

**3** Connect the power cable to the power connector at the rear of the module.

Figure 10 Rearview of Autosampler

- 4 Connect the CAN cable to other Agilent 1260 Infinity modules.
- **5** Connect the APG remote cable (optional) for non-Agilent instruments.
- **6** Turn on the power by pushing the button at the lower left hand side of the module.

The power button stays pressed in and the status LED should be green.

When the line power button stands out and the green light is off, the module is turned off.

# NOTE

NOTE

The module was shipped with default configuration settings. For changing these settings, refer to section *Setting the 8-bit configuration switch*.

# Flow Connections to the Autosampler

Parts required	Description
	System
	Capillaries and tubing from Accessory Kit.
Preparations	Autosampler is installed in system.
NOTE	In an Agilent 1260 Infinity Liquid Chromatograph, the Autosampler is located between a Pump (above) and the Thermostatted Column Compartment (below), see "Optimizing the Stack Configuration" on page 34

# WARNING

### Toxic, flammable and hazardous solvents, samples and reagents

The handling of solvents, samples and reagents can hold health and safety risks.

- → When working with these substances observe appropriate safety procedures (for example by wearing goggles, safety gloves and protective clothing) as described in the material handling and safety data sheet supplied by the vendor, and follow good laboratory practice.
- The volume of substances should be reduced to the minimum required for the analysis.
- → Do not operate the instrument in an explosive atmosphere.
- **1** Open the front cover by pressing the button on the right side of the module.
- **2** Install the capillary from the pump outlet into the port 1 of the injection valve.
- **3** Install the capillary from the port 6 of the injection valve to the TCC.

# NOTE

The Autosampler can only be operated with the front and side covers closed.

Installation of stainless steel cladded PEEK capillaries

# Installation of stainless steel cladded PEEK capillaries

The Agilent 1260 Infinity Bio-inert LC System uses PEEK capillaries, which are cladded with stainless steel, see "Autosampler Accessory Kit Contents" on page 33.



For bio-inert modules use bio-inert parts only!

These capillaries combine the high pressure stability of steel with the inertness of PEEK. They are used in the high pressure flow path after sample introduction (needle seat capillary) through the TCC/heat exchangers to the column. Such capillaries need to be installed carefully in order to keep them tight without damaging them by overtightening.

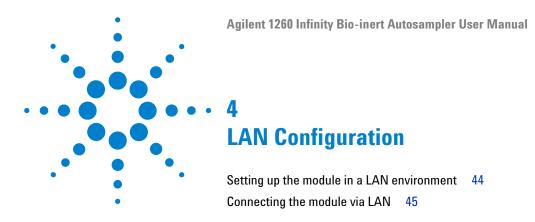
See Agilent 1260 Infinity Bio-inert Quaternary LC System Manual for correct installation.

### CAUTION

Strong force/torque will damage SST cladded PEEK capillaries

Be careful when installing stainless steel cladded PEEK capillaries.

→ See Agilent 1260 Infinity Bio-inert Quaternary LC System Manual for correct installation.



This chapter provides information on connecting the autosampler to the Agilent ChemStation PC.



### 4 LAN Configuration

Setting up the module in a LAN environment

# Setting up the module in a LAN environment

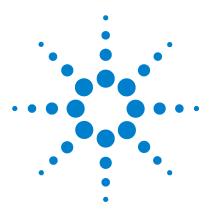
It is not recommended to connect an Agilent 1260 Infinity system via the G5667A Autosampler. The detector is producing the most data in the stack, followed by the pump, and it is therefore highly recommended to use either of these modules for the LAN connection.

# **Connecting the module via LAN**

If the module is being operated as a standalone module or if a connection via LAN is required regardless of above mentioned recommendation, a G1369B/C LAN card has to be used. For installation and configuration, see the G1369B/C documentation.

# 4 LAN Configuration

Connecting the module via LAN



5

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Method Parameter Settings 57
Module Configuration 61
Main Screens of the Autosampler with Agilent Instant Pilot (G4208A) 62

This chapter provides information on how to set up the autosampler for an analysis and explains the basic settings.



# **Preparing the Autosampler**

# **Preparing the Autosampler**

For best performance of the autosampler

- When using the Autosampler in a system with a vacuum degassing unit, shortly degas your samples before using them in the autosampler.
- Filter samples before use in 1260 system. Use High pressure filter kit (p/n 5067-4638) for inline filtering.
- When using buffer solutions, flush the system with water before switching it off.
- Check the autosampler plungers for scratches, grooves and dents when changing the piston seal. Damaged plungers cause micro leaks and will decrease the lifetime of the seal.
- Priming and Purging the System When the solvents have been exchanged or the system has been turned off for a certain time (for example, overnight) oxygen will re-diffuse into the solvent channel. Therefore priming and purging of the system is required before starting an application.

Activity	Solvent	Comments
After an installation	lsopropanol	Best solvent to flush air out of the system
When switching between reverse phase and normal phase (both times)	Isopropanol	Best solvent to flush air out of the system
After an installation	Ethanol or methanol	Alternative to isopropanol (second choice) if no isopropanol is available
To clean the system when using buffers	Bidistilled water	Best solvent to re-dissolve buffer crystals
After a solvent change	Bidistilled water	Best solvent to re-dissolve buffer crystals

 Table 5
 Choice of Priming Solvents for Different Purposes

# **Solvent Information**

Observe the following recommendations on the use of solvents.

- Follow recommendations for avoiding the growth of algae, see pump manuals.
- Small particles can permanently block capillaries and valves. Therefore, always filter solvents through 0.4  $\mu m$  filters.
- Avoid or minimize the use of solvents that may corrode parts in the flow path. Consider specifications for the pH range given for different materials like flow cells, valve materials etc. and recommendations in subsequent sections.

# Solvent information for parts of the 1260 Infinity Bio-inert LC system

For the Agilent 1260 Infinity Bio-inert LC system, Agilent Technologies uses highest quality materials (see "Bio-inert Materials" on page 11) in the flow path (also referred to as wetted parts), which are widely accepted by life scientists, as they are known for optimum inertness to biological samples, and ensure best compatibility to common samples and solvents over a wide pH range. Explicitly, the complete flow path is free from stainless steel and free from other alloys containing metals such as iron, nickel, cobalt, chromium, molybdenum or copper, which can interfere with biological samples. The flow downstream of the sample introduction contains no metals whatsoever.

However, there are no materials that combine suitability for versatile HPLC instrumentation (valves, capillaries, springs, pump heads, flow cells etc.) with complete compatibility with all possible chemicals and application conditions. This section recommends the preferred solvents. Chemicals that are known to cause issues should be avoided, or exposure should be minimized, for example, for short-term cleaning procedures. After potentially aggressive chemicals have been used, the system should be flushed with compatible standard HPLC solvents.

# PEEK

PEEK (Polyether-Ether Ketones) combines excellent properties with regard to biocompatibility, chemical resistance, mechanical and thermal stability and is therefore the material of choice for biochemical instrumentation. It is stable in the specified pH range, and inert to many common solvents. There is still a number of known incompatibilities with chemicals such as chloroform, methylene chloride, THF, DMSO, strong acids (nitric acid > 10 %, sulphuric acid > 10 %, sulfonic acids, trichloroacetic acid), halogenes or aequous halogene solutions, phenol and derivatives (cresols, salicylic acid etc.).

When used above room temperature, PEEK is sensitive to bases and various organic solvents, which can cause it to swell. As normal PEEK capillaries are very sensitive to high pressure, especially under such conditions, Agilent uses stainless-steel cladded PEEK capillaries to keep the flow path free of steel and to ensure pressure stability to at least 600 bar. If in doubt, consult the available literature about the chemical compatibility of PEEK.

### Titanium

Titanium is highly resistant to oxidizing acids (for example, nitric, perchloric and hypochlorous acid) over a wide range of concentrations and temperatures. This is due to a thin oxide layer on the surface, which is stabilized by oxidizing compounds. Reducing acids (for example, hydrochloric, sulfuric and phosphoric acid) can cause slight corrosion, which increases with acid concentration and temperature. For example, the corrosion rate with 3 % HCl (about pH 0.1) at room temperature is about 13  $\mu$ m/year. At room temperature, titanium is resistant to concentrations of about 5 % sulfuric acid (about pH 0.3). The addition of nitric acid to hydrochloric or sulfuric acids significantly reduces corrosion rates. Titanium is subject to corrosion in anhydrous methanol, which can be avoided by adding a small amount of water (about 3 %). Slight corrosion is possible with ammonia > 10 %.

### **Fused silica**

Fused silica is inert against all common solvents and acids except hydrofluoric acid. It is corroded by strong bases and should not be used above pH 12 at room temperature. The corrosion of flow cell windows can negatively affect measurement results. For a pH greater than 12, the use of flow cells with sapphire windows is recommended.

# Gold

Gold is inert to all common HPLC solvents, acids and bases within the specified pH range. It can be corroded by complexing cyanides and concentrated acids like aqua regia (a mixture of concentrated hydrochloric and nitric acid).

# **Zirconium Oxide**

Zirconium Oxide  $(ZrO_2)$  is inert to almost all common acids, bases and solvents. There are no documented incompatibilities for HPLC applications.

# Platinum/Iridium

Platinum/Iridium is inert to almost all common acids, bases and solvents. There are no documented incompatibilities for HPLC applications.

# PTFE

PTFE (polytetrafluorethen) is inert to almost all common acids, bases and solvents. There are no documented incompatibilities for HPLC applications.

# Sapphire, Ruby and Al<sub>2</sub>O<sub>3</sub>-based ceramics

Sapphire, ruby and ceramics based on  $Al_2O_3$  are inert to almost all common acids, bases and solvents. There are no documented incompatibilities for HPLC applications.

Data above were collected from external resources and are meant as a reference. Agilent cannot guarantee the completeness and correctness of such information. Information can also not be generalized due to catalytic effects of impurities like metal ions, complexing agents, oxygen etc. Most data available refers to room temperature (typically 20 - 25 °C, 68 - 77 °F). If corrosion is possible, it usually increases at higher temperatures. If in doubt, consult additional resources.

### Using the Module Setting up the Autosampler with Agilent ChemStation

# Setting up the Autosampler with Agilent ChemStation

The setup of the Autosampler is shown with the Agilent ChemStation B.04.02. SP1 DSP3. Depending on the controller (e.g. Agilent Instant Pilot, EZChrom Elite) the screens look different. For the Instant Pilot refer to "Main Screens of the Autosampler with Agilent Instant Pilot (G4208A)" on page 62.

NOTE

5

This section describes the autosampler settings only. For information on the Agilent ChemStation or other 1260 Infinity modules refer to the corresponding documentation.

Setting up the Autosampler with Agilent ChemStation

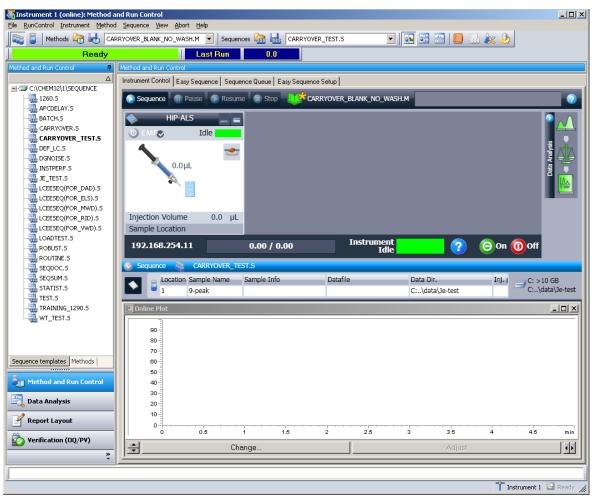
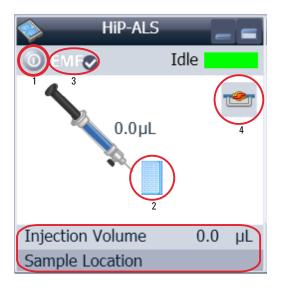


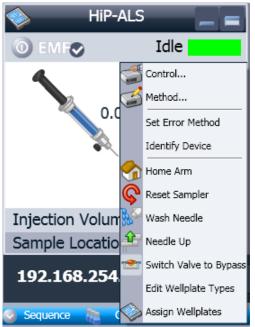
Figure 11 ChemStation Method & Run Control

After successful load of the ChemStation, you should see the module as an active item in the graphical user interface (GUI).

Setting up the Autosampler with Agilent ChemStation



# The Autosampler User Interface



Within the Autosampler user interface, there are active areas. If you move the mouse cursor across the icons (tray, EMF button), the cursor will change and you may click on the icon to

- Turn on/off the autosampler (1)
- Configure the sample tray (2)
- Get the status of the EMF (Early Maintenance Feature) (3)
- Switch injection valve to Mainpass / Bypass (4)

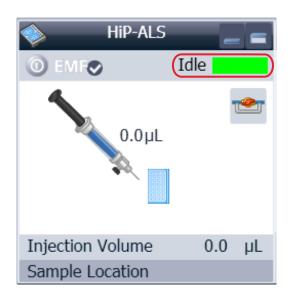
Instrument actuals Information

- Injection volume
- · Sample location

A right-click into the Active Area will open a menu to

- Show the Control User Interface (special module settings)
- Show the Method User interface (same as via menu Instrument – Setup G1367E)
- Set Error Method
- Identify Device
- Home Arm
- Reset Sampler
  - Wash Needle
- Needle Up
- Valve Mainpass / Bypass (same as click on the valve icon)
- Switch on Tray Illumination
- Edit Well Plate Types
- Wellplate Configuration (same as click on the Tray icon)

Setting up the Autosampler with Agilent ChemStation



# HiP-ALS EMF Idle 0.0μL 0.0μL Injection Volume 0.0 μL Sample Location

# Module Status shows Run / Ready / Error state and "Not Ready text" or "Error text".

- Error (Red)
  Not ready (r
- Not ready (yellow)
- Ready (green)
- Pre run, Post run (purple)
- Run (blue)
- Idle (green)
- Offline (dark gray)
- · Standby (light gray)

**EMF Status** shows Run / Ready / Error state and "Not Ready text" or "Error text".

- Offline (gray)
- Ok. No Maintenance required (green)
- EMF warning. Maintenance might be required (yellow)
- EMF warning. Maintenance required (red)

Setting up the Autosampler with Agilent ChemStation

# **Control Settings**

Control		×
Missing Vessel		
	Ignore missing vessel	
Linked Pump		
	G4220A:DE92900137	•
Prime Flush Pump		
	۲	off
	Ô	on for 5 🔅 sec
	<u> </u>	<u>C</u> ancel Help

These settings are available via right click on the Active Area of the ALS GUI.

**Missing Vessel**: The handling of missing vessels can be configured.

**Linked Pump**: To configure which pump delivers flow to the Autosampler.

Prime Flush Pump: Priming the Needle wash flush pump.

# **Method Parameter Settings**

These settings are available via **Menu > Instrument > Setup Agilent 1260 Infinity Autosampler** or via right click on the Active area.

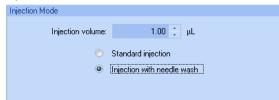
# NOTE

The signal window in the lower part is not shown when opening the parameter settings via right mouse on the Autosampler user interface.

Method of G5667A (LP00055727)			_ D
		HiP Sampler (G5667A)	
Injection Mode	+ Advanced		
Injection volume: 5.00 🛟 µL	Auxiliary		
Standard injection	Draw speed:	200.0 📜 µL/min	
Injection with needle wash	Eject speed:	200.0 🌻 µL/min	
	Draw position:	0.0 ; mm	
Needle wash	Equilibration time:	2.0 📜 sec	
	Sample flush out factor:	5.0 📜 times injection volume	
Mode: Flush Port		Vial/Well bottom sensing	
Time: 3.0 ; sec			
Location:	High throughput		
Repeat 3 📫 times	Automat	ic delay volume reduction	
toptime Posttime	🗖 Enable o	overlapped injection	
	O W	/hen Sample Is Flushed Out	
As Pump/No Limit     Off	O Af	ter Period Of Time	
© <u>1.00 ;</u> min © <u>1.00 ;</u> min		0.00 📜 min	
	< ]	1	>
	🛨 Injection Cleaning		
		Ok	Cancel

Setting up the Autosampler with Agilent ChemStation

### **Injection Mode**



The settable **Injection volume** range is from  $0.1 - 20.0 \mu L$ . Select to use **Standard injection** or **Injection with Needle wash**.

### Needle wash

Needle wash		
Mode:	Flush Port	¥
Time:	3.0 ÷	sec
Location:		
Repeat:	3 🛟	times

It is possible to select between using the built in flush port of the Autosampler or using a non-capped vial. Using **needle wash** is required to obtain minimum carry-over.

### **Stop Time**

Stoptime				
~				
۲	No Limit			
0		1.00	÷	min

An autosampler Stop Time can be set.

### **Injection Cleaning**

Injection Cleaning		
Injection Valve Cleaning		
Time 1: 🔽	0.01 🗘	min (Bypass)
Time 2: 🗾	0.01	min (Mainpass/Bypass)
Time 3: 🕅	0.01	min (Mainpass/Bypass)
Time 4: 🛄	0.01	min (Mainpass/Bypass)
Valve movements:	0 :	

The **Injection Valve Cleaning** section allows you to specify the valve switching times at the end of overlap or sample flush.

Times 1 ... 4 are the times when the valve switches to bypass (for time 1) or to mainpass and bypass (for times 2, 3 and 4).The times must be specified in ascending order. You can also switch the times to off. Between the first and second, and second and third valve switches, a rinse is executed using the rinse volumes specified in the Injector Cleaning section.

**Valve movements** specifies the number of times that the valve switches from mainpass to bypass at times 2, 3 and 4 in the field.The maximum value is 2; default is 1.

5

Setting up the Autosampler with Agilent ChemStation

Set	up Method					×		
Ś	IIP-ALS 🗇 HIP-ALS Injector Program							
R	Iv Use Injector Program							
Function Parameter								
	Draw	-	Draw del	ault volume	from sample wit	h default spee		
1	Append		sert	Delete	Clear all	Move up		
		_						
	Cut		ору	Paste		Move <u>d</u> own		
	OK		Apply		Cancel	Help		

### **Injection Program**

The pretreatment/injector program comprises a series of numbered lines, each specifying an operation that the autosampler carries out sequentially. When you activate a pretreatment/injector program, it replaces the standard injection cycle.

Select **Append** to add the contents of the edit line to the end of the table.

Select **Insert** to insert the contents of the edit line above the currently-selected line.

Select **Delete** to delete the currently selected line.

Select **Clear All** to clear all pretreatment/injector program functions from the table.

Select **Move up** to move the currently selected line one position up in the order of execution.

Select **Move down** to move the currently selected line one position down in the order of execution.

Select **Cut** to delete the currently-selected line and place it on the clipboard.

Select **Copy** to copy the currently selected line to the clipboard.

Select **Paste** to paste the line on the clipboard at the current position.

# **Module Configuration**

iP Autosampler Configuration: Instrument 1	×						
Communication	U						
Device name HiP Sampler							
Type ID G5667A 👻							
Serial number LP00055727							
Firmware revision T.06.33 [004]							
Connection settings							
Options	1						
Syringe 100 💌 µL							
Seat Capillary 2.3 🗸 µL							
Max. injection volume 100.00 µL							
External contacts board installed							
use BCD port for							
Cocation O Binary Output							
BCD port output format							
BCD O Binary							
Thermostat installed							
Flexible Cube installed							
Rinse valve installed							
Rinse valve enabled							
Define Wellplates							
OK Cancel Help							

These settings are available via menu Instrument > More 1260 Infinity ALS > Autosampler Configuration.

Device name: based on the module. Type ID: based on the module (product number). Some modules may allow changing the type based on hardware/firmware. This results in a change of features and functions. Serial number: based on the module. Firmware revision: based on the module.

Options: lists installed options.

Main Screens of the Autosampler with Agilent Instant Pilot (G4208A)

# Main Screens of the Autosampler with Agilent Instant Pilot (G4208A)

Welcome						
				Control Details		
1260 lr	finity	HiP AL G5667/	.S SL A - LP00055727	09:41		
Method	Sequence	Status	Logbook	More 🛆		

Below the main screens for the use of the detector are shown.

### The Control screen allows

- System: On/Off
- · System: Get Ready
- System: Clear Errors
- HIP ALS: Wash needle

Property	Value
Resident Revison	B.06.32 [0004]
On-time	9d 23:36h Reload
Installed Options	Seal Wash Pump, Solvent Selection Valve, Bootp
LAN TCP/IP Mode	BOOTP
LAN TCP/IP Address	0.0.0.0 Print
LAN MAC Address	0030D31BD38F
Board ID	TYPE=G4220-65800, REV=AK, MFG=ZZ, SER=M/
Hi	P ALS SL : LP00055727
Main Revision	T.06.33 [004]
Resident Revison	A.06.10 [004]
On-time	0d 00:00h
Board ID	TYPE="G4226-65000", REV="CA_AA", SER="0107
Syringe	:
	DAD : DEBAF00163
Main Revision	B.06.32 [0004]
Resident Revison	B.06.32 [0004]
On-time	9d 23:37h Exit

### The System Info screen lists details of the autosampler

- Firmware revision
- On-time
- Main Board information
- Transport assembly information
- · Sampling unit information
- · Syringe information

Main Screens of the Autosampler with Agilent Instant Pilot (G4208A)

	Configure - HiP ALS	
		🛃
		Edit
Setting	Value	
Symbolic Name	<not set=""></not>	
Volumes	Syringe 20 µl, Seat 1.2 µl	Plates
On Missing Vessel	Abort	
Plate 1 (Front)	<no plate=""></no>	
Plate 2 (Back)	<no plate=""></no>	
Flush-Out Pump	BINPUMP	
Serial Interface	19200 Baud, 8 Bits, None Parity	
Sample Illumination	OFF	
	<b>v</b>	Exit
-		
A user-defined identifier f	or the module.	14:02
System Contro	ller 🛛 Bin Pump 🔹 HiP ALS	Þ
Me Me	ethod - UNNAMED* filtered	

### The **Configure** screen allows to configure

- Symbolic Name of module
- Volumes
- On Missing Vessel behaviour
- Plate configuration
- Flush-Out Pump
- Serial Interface configuration
- Sample Illumination

The **Method** screen lists all method parameters of the autosampler. These can be edited.

Setting	Valu	le		🐺
	S	vstem		Edit
	BINPUMP	: LP00000005		
	HIP ALS	: PP00055050		
<b>Injection Volu</b>	me 1.00	μΙ		Control
Injection Mod	e Stan	dard		
Overlap	Disa	bled		
Needle Wash	3.0 s	ec in Flush Por	t	- I I
Wash Position	ע V 10			Toggle
Draw Speed	100.	0 µl/min		_
Eject Speed	100.	0 µl/min		_
	TCC : E	E00000009		
	DAD : D	DE00000000		
				<u>▼</u> Exit
				14:11
Filter	Compare	Timetable	Properties	File

Main Screens of the Autosampler with Agilent Instant Pilot (G4208A)

	Maintenance - HiP ALS	
Maaaaaa	Date Time	EXF
Message	EMF Events	- V I
[Empty]		Setup
	Error Events	
[Empty]	Maintenance Entries	Maint.
[Empty]	Maintenance Entries	
[=,p+3]		🕱
		Entry
		Ident.
		Ident.
-		
ļ	• • •	14:03
System	Controller Bin Pump HiP ALS	Þ
	Diagnosis	
	Diagnosis	
	Diagnosis BINPUMP : LP00000005	
Pressure Test	BINPUMP : LP0000005	
Pressure Test	BINPUMP : LP00000005	 Exec.
	BINPUMP : LP00000005	
Pressure Test	BINPUMP : LP00000005	
Pressure Test	BINPUMP : LP00000005	
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	Exec.
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	Exec.
Pressure Test Injector Steps Lamp Intensit Calibration Te	BINPUMP : LP00000005	Exec.

### The Maintenance screen allows

- EMF setup
- logging of maintenance activities

• module identification (blinking LED)

Firmware updates can be done via the System Maintenance screen.

The **Diagnosis** screen provides access to module specific tests.

• Injector steps



# **Optimizing Performance**

Delay Volume and Extra-Column Volume 66
Delay Volume 66
How to Configure the Optimum Delay Volume 67
How to Achieve Higher Injection Volumes 69
How to Achieve High Throughput 71
How to Achieve Higher Resolution 72
How to Achieve Higher Sensitivity 75
How to Achieve Lowest Carry Over 76

This chapter gives hints on how to optimize the performance or use additional devices.



# **Delay Volume and Extra-Column Volume**

The *delay volume* is defined as the system volume between the point of mixing in the pump and the top of the column.

The *extra-column volume* is defined as the volume between the injection point and the detection point, excluding the volume in the column.

# **Delay Volume**

In gradient separations, this volume causes a delay between the mixture changing in the pump and that change reaching the column. The delay depends on the flow rate and the delay volume of the system. In effect, this means that in every HPLC system there is an additional isocratic segment in the gradient profile at the start of every run. Usually the gradient profile is reported in terms of the mixture settings at the pump and the delay volume is not quoted even though this will have an effect on the chromatography. This effect becomes more significant at low flow rates and small column volumes and can have a large impact on the transferability of gradient methods. It is important, therefore, for fast gradient separations to have small delay volumes, especially with narrow bore columns (e.g., 2.1 mm i.d.) as often used with mass spectrometric detection.

The delay volume in a system includes the volume in the pump from the point of mixing, connections between pump and autosampler, volume of the flow path through the autosampler and connections between autosampler and column.

# How to Configure the Optimum Delay Volume

For very fast gradients over 0.5 min the delay volume of the system can be easily reduced without changing the physical configuration of the system. The change is achieved by changing the behavior of the autosampler.

The 270  $\mu$ L delay volume of the autosampler is due to the flow path from the injection valve through the metering device, needle, needle seat and connecting capillaries back to the injection valve (see Figure 12 on page 68). To make an injection the valve switches from mainpass to bypass so that the metering device can draw the sample into the needle capillary. The injection is made when the valve switches back to mainpass and the sample is flushed onto the column. The valve remains in this position during analysis so that the autosampler is continually flushed and hence the gradient has to flow through this delay volume to reach the column. This can be eliminated by switching the injection valve from mainpass to bypass after the injection has been made and the injected sample has been flushed onto the column. In practice this can be done a few seconds after injection and is activated by selecting the "Automatic Delay Volume Reduction" (ADVR) function in the autosampler setup menu. The Flush-out Factor (typically 5 times injection volume) ensures that enough time is allowed to flush the sample out of the injector before switching to bypass. For instance a 1 µL injection under standard conditions effectively reduces the system delay volume by approximatly 250 µL.

### **6** Optimizing Performance

How to Configure the Optimum Delay Volume

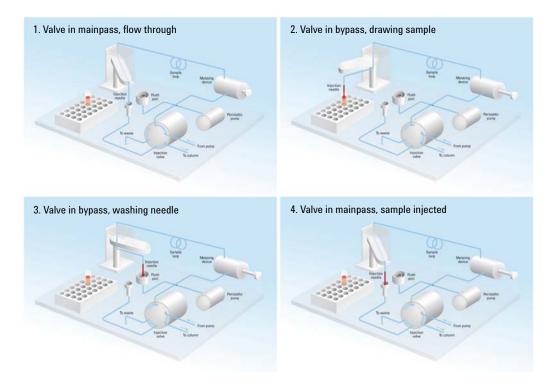


Figure 12 Schematic of injection steps in 1260 Infinity Autosampler

When using ADVR it should be noted that the gradient has already started at the pump at the instant of injection. The question should be asked whether the gradient has already reached the autosampler, in which case a small step in the gradient will result. This happens when the delay volume is less than the flush-out volume and is not necessarily a problem but may be a factor to be considered in a method transfer. With a flush-out factor of 5 and an injection volume of 10  $\mu$ L, the autosampler will allow 50  $\mu$ L to pass through before switching to bypass which, with a delay volume of 50  $\mu$ L, means the gradient just reached the injection valve. Smaller injection volumes will have no effect but for larger injection volumes this will introduce a small step in the gradient. The flow rate in use will also have an impact on the decision to use ADVR or not. At 0.2 mL/min the delay time saved is 21 seconds while at 1.0 mL/min it is 4 seconds.

The ADVR function is unlikely to be suitable for applications involving compounds which are known to cause carry-over problems.

# How to Achieve Higher Injection Volumes

The standard configuration of the Agilent 1260 Infinity Bio-inert Autosampler can inject a maximum volume of 100  $\mu$ l with the bio-inert loop capillary. To increase the injection volume the Multidraw upgrade kit (p/n G5667-68711) can be installed. With the kit you can add a maximum of 400  $\mu$ l or 1400  $\mu$ l to the injection volume of your injector. The total volume is then 500  $\mu$ l or 1500  $\mu$ l for the autosampler with 100  $\mu$ l analytical head. Note the delay volume of your autosampler is extended when using the extended seat capillaries from the multi-draw kit. When calculating the delay volume of the autosampler you have to double the volume of the extended capillaries. The delay volume can be reduced by bypassingthe autosampler once the sample has reached the head of the column. The system delay volume due to the autosampler will increase accordingly.

Whenever a method is scaled down from a larger column to a smaller column it is important that the method translation makes an allowance for reducing the injection volume in proportion to the volume of the column to maintain the performance of the method. This is to keep the volume of the injection at the same percentage volume with respect to the column. This is particular important if the injection solvent is stronger (more eluotropic) than the starting mobile phase and any increase will affect the separation particularly for early running peaks (low retention factor). In some cases it is the cause of peak distortion and the general rule is to keep the injection solvent the same or weaker than the starting gradient composition. This has a bearing on whether, or by how much, the injection volume can be increased and the user should check for signs of increased dispersion (wider or more skewed peaks and reduced peak resolution) in trying to increase the injection size. If an injection is made in a weak solvent then the volume can probably be increased further because the effect will be to concentrate the analyte on the head of the column at the start of the gradient. Conversely if the injection is in a stronger solvent than the starting mobile phase then increased injection volume will spread the band of analyte down the column ahead of the gradient resulting in peak dispersion and loss of resolution.

Perhaps the main consideration in determining injection volume is the diameter of the column as this will have a big impact on peak dispersion. Peak heights can be higher on a narrow column than with a larger injection on a wider column because there is less peak dispersion. With 2.1 mm i.d. columns

**How to Achieve Higher Injection Volumes** 

typical injection volumes might range up to 5 to10  $\mu$ l but it is very dependent on the chemistry of the analyte and mobile phase as discussed above. In a gradient separation injection volumes of about 5 % of the column volume might be achieved whilst maintaining good resolution and peak dispersion.

One way to achieve larger injections is to use a trapping column selected by a switching valve to capture and concentrate the injection before switching it, i.e. injecting it, onto an analytical column, see Figure 13 on page 70. The valve can be conveniently located in the Thermostatted Column Compartment.

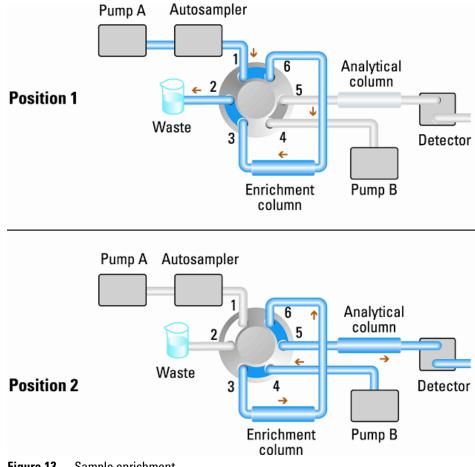


Figure 13 Sample enrichment

# How to Achieve High Throughput

The injection can be optimized for speed remembering that drawing the sample too fast can reduce the reproducibility. Marginal gains are to be made here as the sample volumes used tend towards the smaller end of the range in any case. A significant portion of the injection time is the time taken with the needle movements to and from the vial and into the flush port. These manipulations can be performed while the previous separation is running. This is known as "overlapped injection" and it can be easily turned on from the autosampler setup screen in the ChemStation Control Software. The autosampler can be told to switch the flow through the autosampler to bypass after the injection has been made and then after, for example, 3 minutes into a 4 minutes run to start the process of aspirating the next sample and preparing for injection. This can typically save 0.5 to 1 minute per injection.

# **How to Achieve Higher Resolution**

Increased resolution in a separation will improve the qualitative and quantitative data analysis, allow more peaks to be separated or offer further scope for speeding up the separation. This section considers how resolution can be increased by examining the following points:

- Optimize selectivity
- Smaller particle-size packing
- Longer Columns
- · Shallower gradients, faster flow

Resolution between two peaks is described by the resolution equation:

$$Rs = \frac{1}{4}\sqrt{N}\frac{(\alpha - 1)}{\alpha}\frac{(k_2 + 1)}{k_2}$$

where

- R<sub>s</sub>=resolution,
- N=plate count (measure of column efficiency),
- *α*=selectivity (between two peaks),
- k<sub>2</sub>=retention factor of second peak (formerly called capacity factor).

The term that has the most significant effect on resolution is the selectivity,  $\alpha$ , and practically varying this term involves changing the type of stationary phase (C18, C8, phenyl, nitrile etc.), the mobile phase and temperature to maximize the selectivity differences between the solutes to be separated. This is a substantial piece of work which is best done with an automated method development system which allows a wide range of conditions on different columns and mobile phases to be assessed in an ordered scouting protocol. This section considers how to get higher resolution with any chosen stationary and mobile phases. If an automated method development system was used in the decision on phases it is likely that short columns were used for fast analysis in each step of the scouting.

The resolution equation shows that the next most significant term is the plate count or efficiency, N, and this can be optimized in a number of ways. N is inversely proportional to the particle size and directly proportional to the length of a column and so smaller particle size and a longer column will give a higher plate number. The pressure rises with the inverse square of the particle size and proportionally with the length of the column. This is the reason that the 1260 Infinity LC system was designed to go to 600 bar so that it can run sub-two-micron particles and column length can be increased to 100 mm or 150 mm. There are even examples of 100 mm and 150 mm columns linked to give 250 mm length. Resolution increases with the square root of N so doubling the length of the column will increase resolution by a factor of 1.4. What is achievable depends on the viscosity of the mobile phase as this relates directly to the pressure. Methanol mixtures will generate more back pressure than acetonitrile mixtures. Acetonitrile is often preferred because peak shapes are better and narrower in addition to the lower viscosity but methanol generally yields better selectivity (certainly for small molecules less than about 500 Da). The viscosity can be reduced by increasing the temperature but it should be remembered that this can change the selectivity of the separation. Experiment will show if this leads to increase or decrease in selectivity. As flow and pressure are increased it should be remembered that frictional heating inside the column will increase and that can lead to slightly increased dispersion and possibly a small selectivity change both of which could be seen as a reduction in resolution. The latter case might be offset by reducing the temperature of the thermostat by a few degrees and again experiment will reveal the answer.

The van Deemter curve shows that the optimum flow rate through an STM column is higher than for larger particles and is fairly flat as the flow rate increases. Typical, close to optimum, flow rates for STM columns are: 2 ml/min for 4.6 mm i.d.; and 0.4 ml/min for 2.1 mm i.d. columns.

#### **6** Optimizing Performance

**How to Achieve Higher Resolution** 

In isocratic separations, increasing the retention factor, k, results in better resolution because the solute is retained longer. In gradient separations the retention is described by  $k^*$  in the following equation:

$$k^* = \frac{t_G}{\Delta\%B} \cdot \frac{F}{V_m} \cdot \frac{100}{S}$$

where:

- $k^*$  = mean k value,
- t<sub>G</sub> = time length of gradient (or segment of gradient) (min),
- F = flow (ml/min),
- V<sub>m</sub> = column delay volume,
- $\Delta$ %B = change in fraction of solvent B during the gradient,
- S = constant (ca. 4-5 for small molecules).

This shows that k and hence resolution can be increased by having a shallower gradient (2 to 5 %/min change is a guideline), higher flow rate and a smaller volume column. This equation also shows how to speed up an existing gradient

- if the flow is doubled but the gradient time is halved, k<sup>\*</sup> remains constant and the separation looks the same but happens in half the time. Recently published research has shown how a shorter STM column (at temperatures above 40 °C) can generate higher peak capacity than a longer STM column by virtue of running it faster. (Refer to *Petersson et al., J.Sep.Sci, 31, 2346-2357, 2008, Maximizing peak capacity and separation speed in liquid chromatography*).

## How to Achieve Higher Sensitivity

The sensitivity of a separation method is linked to the choice of stationary and mobile phases as good separation with narrow peaks and a stable baseline with minimal noise are desirable. The choice of instrument configuration will have an effect and a major impact is the setup of the detector. This section considers how sensitivity is affected by:

- Pump mixer volume
- Narrower columns
- Detector flow cell
- Detector parameters

In addition, the discussion on detector parameters also mentions the related topics of selectivity and linearity.

#### Columns

Sensitivity is specified as a signal-to-noise ratio (S/N) and hence the need to maximize peak height and minimize baseline noise. Any reduction in peak dispersion will help to maintain peak height and so extra-column volume should be minimized by use of short, narrow internal diameter, connection capillaries and correctly installed fittings. Using smaller inner diameter columns should result in higher peak height and is therefore ideal for applications with limited sample amounts. If the same sample amount can be injected on a smaller i.d. column, then the dilution due to column diameter will be less and the sensitivity will increase. For example, decreasing the column i.d. from 4.6 mm to 2.1 mm results in a theoretical gain in peak height of 4.7 times due to the decreased dilution in the column. For a mass spectrometer detector, the lower flow rates of narrow columns can result in higher ionization efficiencies and therefore higher sensitivity.

## How to Achieve Lowest Carry Over

Carryover is measured when residual peaks from a previous active-containing injection appear in a subsequent blank solvent injection. There will be carry over between active injections which may lead to erroneous results. The level of carryover is reported as the area of the peak in the blank solution expressed as a percentage of the area in the previous active injection. The Agilent 1260 Infinity autosampler is optimized for lowest carryover by careful design of the flow path and use of materials in which sample adsorption is minimized. A carryover figure of 0.002 % should be achievable even when a triple quadrupole mass spectrometer is the detector. Operating settings of the autosampler allow the user to set appropriate parameters to minimize carryover in any application involving compounds liable to stick in the system.

The following functions of the autosampler can be used to minimize carryover:

- Internal needle wash
- External needle wash
- Needle seat backflush
- Injection valve cleaning

The flow path, including the inside of the needle, is continuously flushed in normal operation, providing good elimination of carryover for most situations. Automated delay volume reduction (ADVR) will reduce the delay volume but will also reduce the flushing of the autosampler and should not be used with analytes where carryover might be a problem.

The outside of the needle can be washed using a wash vial in a specific location or the needle can be washed using the flush port. If a wash vial in a tray location specified by the user is chosen then this vial should have no septum and should contain a solvent suitable for washing the sample from the needle. The septum is not used to avoid wiping contamination off the needle on the downstream only to re-apply it on the upstroke. The needle can be dipped into the vial multiple times. This will be effective in removing a small degree of carryover but for more effective washing of the outside of the needle use the flushport.

The flush port is located above and behind the needle seat and a peristaltic pump delivers the wash solvent. It has a volume of 0.68 ml and the peristaltic pump delivers 6 ml/min, which means the flush port volume is completely

refilled with fresh solvent in 7 s. If the flush port is selected, the user can set how long the outside of the needle is to be washed with fresh solvent. This may be as low as two or three seconds in routine situations where carryover is less of a problem and 10 to 20 s for more complete washing. It is recommended that washing the outside of the needle in the flush port should be standard procedure to avoid contaminating the needle seat. If the needle seat becomes contaminated it will have to be back-flushed, by manually changing the flow connections, to clean it. This is one of the tasks that can be automated using the Flexible Cube module.

The flush port and its solvent delivery pump and tubing should be regularly flushed to ensure the lowest carryover. For example, before using the system each day, prime the flush pump for three minutes with appropriate solvent.

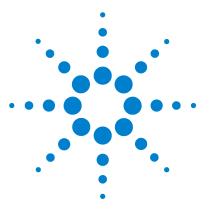
When other measures have failed to eliminate carryover it might be that analyte is sticking inside the injector valve. The injector valve can be set to make additional switching movements to clean out the flow path in the valve if problems occur here with carryover. If the problem compounds need a high percentage of organic phase for elution, it is recommended to switch the injection valve at the high percentage of organic phase after the last peak has eluted. It is also recommended to switch the injection valve again after the initial conditions for the mobile phase have stabilized. This ensures that the bypass groove in the rotor seal of the valve contains the gradient start conditions, which is especially important for flow rates below 0.5 ml/min.

For samples where the outside of the needle cannot be cleaned sufficiently with water or alcohol from the flush pump use wash vials with an appropriate solvent. With an injector program several wash vials can be used for cleaning.

The optimum carry-over performance of the Autosampler is achieved after a run-in period of new instruments or after the exchange of consumable parts (like needle, needle seat and valve parts). During injections in this period, surfaces of these parts adjust to each other. After this period, we recommend back-flushing the needle seat in order to get the sealing areas between needle and needle seat clean. Regular Preventive Maintenance service is recommended as the carry-over performance of the Autosampler depends on the integrity of these consumable parts. Using the G4227A Flexible Cube will additionally improve the carry-over performance and life time of these parts.

### **6** Optimizing Performance

How to Achieve Lowest Carry Over



7

# **Troubleshooting and Diagnostics**

Overview of the Module's Indicators and Test Functions 80 Status Indicators 81 Power Supply Indicator 81 Module Status Indicator 82 User Interfaces 83 Agilent Lab Advisor Software 84

This chapter gives an overview about the troubleshooting and diagnostic features and the different user interfaces.



## **Overview of the Module's Indicators and Test Functions**

### **Status Indicators**

7

The module is provided with two status indicators which indicate the operational state (prerun, run, and error states) of the module. The status indicators provide a quick visual check of the operation of the module.

### **Error Messages**

In the event of an electronic, mechanical or hydraulic failure, the module generates an error message in the user interface. For each message, a short description of the failure, a list of probable causes of the problem, and a list of suggested actions to fix the problem are provided (see chapter Error Information).

## **Test Functions**

A series of test functions are available for troubleshooting and operational verification after exchanging internal components (see Tests and Calibrations).

## **Status Indicators**

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	ti	

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

Figure 14 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*.

## **Module Status Indicator**

The module status indicator indicates one of six possible module conditions:

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

If the error occurs during analysis, it is propagated within the LC system, i.e. a red LED may indicate a problem of a different module. Use the status display of your user interface for finding the root cause/module of the error.

- A *blinking* indicator indicates that the module is in resident mode (e.g. during update of main firmware).
- A *fast blinking* indicator indicates that the module is in a low-level error mode. In such a case try to re-boot the module or try a cold-start (see "Special Settings" on page 191. Then try a firmware update (see "Replacing the Module Firmware" on page 151). If this does not help, a main board replacement is required.

## **User Interfaces**

- Depending on the user interface, the available tests and the screens/reports may vary.
- Preferred tool should be Agilent Lab Advisor Software, see "Agilent Lab Advisor Software" on page 84.
- The Agilent ChemStation B.04.02 and above do not include any maintenance/test functions.
- Screenshots used within these procedures are based on the Agilent Lab Advisor Software.

7 Troubleshooting and Diagnostics Agilent Lab Advisor Software

## **Agilent Lab Advisor Software**

The Agilent Lab Advisor software is a standalone product that can be used with or without data system. Agilent Lab Advisor software helps to manage the lab for high quality chromatographic results and can monitor in real time a single Agilent LC or all the Agilent GCs and LCs configured on the lab intranet.

Agilent Lab Advisor software provides diagnostic capabilities for all Agilent 1200 Infinity Series modules. This includes diagnostic capabilities, calibration procedures and maintenance routines for all the maintenance routines.

The Agilent Lab Advisor software also allows users to monitor the status of their LC instruments. The Early Maintenance Feedback (EMF) feature helps to carry out preventive maintenance. In addition, users can generate a status report for each individual LC instrument. The tests and diagnostic features as provided by the Agilent Lab Advisor software may differ from the descriptions in this manual. For details refer to the Agilent Lab Advisor software help files.

The Instrument Utilities is a basic version of the Lab Advisor with limited functionality required for installation, use and maintenance. No advanced repair, troubleshooting and monitoring functionality is included.



# **Error Information**

What are Error Messages 87 General Error Messages 88 Timeout 88 Shutdown 89 Remote Timeout 89 Lost CAN Partner 90 Leak 90 Leak Sensor Open 91 Leak Sensor Short 91 **Compensation Sensor Open** 92 **Compensation Sensor Short** 92 Fan Failed 93 Module Error Messages 94 Exhaust Fan Failed 94 Front Door Error 95 Side Door Error 95 Arm Movement Failed or Arm Movement Timeout 96 Valve to Bypass Failed 97 Valve to Mainpass Failed 98 Needle Lock Failed 99 Needle to Needle Seat Position 100 Needle Carrier Failed 101 Missing Vial or Missing Wash Vial 102 Initialization Failed 103 Metering Home Failed 104 Motor Temperature 105 Invalid Vial Position 106 Peristaltic Pump Error 107



#### 8 Error Information

Agilent Lab Advisor Software

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This chapter describes the meaning of error messages, and provides information on probable causes and suggested actions how to recover from error conditions.

## What are Error Messages

Error messages are displayed in the user interface when an electronic, mechanical, or hydraulic (flow path) failure occurs which requires attention before the analysis can be continued (for example, repair, or exchange of consumables is necessary). In the event of such a failure, the red status indicator at the front of the module is switched on, and an entry is written into the module logbook.

## **General Error Messages**

General error messages are generic to all Agilent series HPLC modules and may show up on other modules as well.

### Timeout

#### Error ID: 0062

The timeout threshold was exceeded.

#### **Probable cause**

- The analysis was completed successfully, and the timeout function switched off the module as requested.
- 2 A not-ready condition was present during a sequence or multiple-injection run for a period longer than the timeout threshold.

#### **Suggested actions**

Check the logbook for the occurrence and source of a not-ready condition. Restart the analysis where required.

Check the logbook for the occurrence and source of a not-ready condition. Restart the analysis where required.

### Shutdown

#### Error ID: 0063

An external instrument has generated a shutdown signal on the remote line.

The module continually monitors the remote input connectors for status signals. A LOW signal input on pin 4 of the remote connector generates the error message.

Probable cause		Suggested actions
1	Leak detected in another module with a CAN connection to the system.	Fix the leak in the external instrument before restarting the module.
2	Leak detected in an external instrument with a remote connection to the system.	Fix the leak in the external instrument before restarting the module.
3	Shut-down in an external instrument with a remote connection to the system.	Check external instruments for a shut-down condition.

## **Remote Timeout**

#### Error ID: 0070

A not-ready condition is still present on the remote input. When an analysis is started, the system expects all not-ready conditions (for example, a not-ready condition during detector balance) to switch to run conditions within one minute of starting the analysis. If a not-ready condition is still present on the remote line after one minute the error message is generated.

Probable cause		Suggested actions
1	Not-ready condition in one of the instruments connected to the remote line.	Ensure the instrument showing the not-ready condition is installed correctly, and is set up correctly for analysis.
2	Defective remote cable.	Exchange the remote cable.
3	Defective components in the instrument showing the not-ready condition.	Check the instrument for defects (refer to the instrument's documentation).

## **Lost CAN Partner**

#### Error ID: 0071

During an analysis, the internal synchronization or communication between one or more of the modules in the system has failed.

The system processors continually monitor the system configuration. If one or more of the modules is no longer recognized as being connected to the system, the error message is generated.

Probable cause		Suggested actions	
1	CAN cable disconnected.	Ensure all the CAN cables are connected correctly.	
		• Ensure all CAN cables are installed correctly.	
2	Defective CAN cable.	Exchange the CAN cable.	
3	Defective main board in another module.	Switch off the system. Restart the system, and determine which module or modules are not recognized by the system.	

### Leak

#### Error ID: 0064

A leak was detected in the module.

The signals from the two temperature sensors (leak sensor and board-mounted temperature-compensation sensor) are used by the leak algorithm to determine whether a leak is present. When a leak occurs, the leak sensor is cooled by the solvent. This changes the resistance of the leak sensor which is sensed by the leak-sensor circuit on the main board.

Pr	obable cause	Suggested actions
1	Loose fittings.	Ensure all fittings are tight.
2	Broken capillary.	Exchange defective capillaries.

## Leak Sensor Open

#### Error ID: 0083

The leak sensor in the module has failed (open circuit).

The current through the leak sensor is dependent on temperature. A leak is detected when solvent cools the leak sensor, causing the leak-sensor current to change within defined limits. If the current falls outside the lower limit, the error message is generated.

Probable cause		Suggested actions
1	Leak sensor not connected to the main board.	Please contact your Agilent service representative.
2	Defective leak sensor.	Please contact your Agilent service representative.
3	Leak sensor incorrectly routed, being pinched by a metal component.	Please contact your Agilent service representative.

## **Leak Sensor Short**

#### Error ID: 0082

The leak sensor in the module has failed (short circuit).

The current through the leak sensor is dependent on temperature. A leak is detected when solvent cools the leak sensor, causing the leak sensor current to change within defined limits. If the current increases above the upper limit, the error message is generated.

Probable cause		Suggested actions
1	Defective leak sensor.	Please contact your Agilent service representative.
2	Leak sensor incorrectly routed, being pinched by a metal component.	Please contact your Agilent service representative.

## **Compensation Sensor Open**

#### Error ID: 0081

The ambient-compensation sensor (NTC) on the main board in the module has failed (open circuit).

The resistance across the temperature compensation sensor (NTC) on the main board is dependent on ambient temperature. The change in resistance is used by the leak circuit to compensate for ambient temperature changes. If the resistance across the sensor increases above the upper limit, the error message is generated.

#### Probable cause

#### Suggested actions

1 Defective main board.

Please contact your Agilent service representative.

## **Compensation Sensor Short**

#### Error ID: 0080

The ambient-compensation sensor (NTC) on the main board in the module has failed (short circuit).

The resistance across the temperature compensation sensor (NTC) on the main board is dependent on ambient temperature. The change in resistance is used by the leak circuit to compensate for ambient temperature changes. If the resistance across the sensor falls below the lower limit, the error message is generated.

Pr	obable cause	Suggested actions
1	Defective main board.	Please contact your Agilent service representative.

## **Fan Failed**

#### Error ID: 0068

The cooling fan in the module has failed.

The hall sensor on the fan shaft is used by the main board to monitor the fan speed. If the fan speed falls below a certain limit for a certain length of time, the error message is generated.

Depending on the module, assemblies (e.g. the lamp in the detector) are turned off to assure that the module does not overheat inside.

Probable cause		Suggested actions
1	Fan cable disconnected.	Please contact your Agilent service representative.
2	Defective fan.	Please contact your Agilent service representative.
3	Defective main board.	Please contact your Agilent service representative.

8 Error Information Module Error Messages

## **Module Error Messages**

These errors are autosampler specific.

## **Exhaust Fan Failed**

#### Error ID: 4456, 4457

The exhaust fan in the module has failed.

The hall sensor on the fan shaft is used by the main board to monitor the fan speed. If the fan speed falls below a certain value the error message is generated and the module shuts down.

Probable cause		Suggested actions
1	Fan cable disconnected.	Please contact your Agilent service representative.
2	Defective fan.	Please contact your Agilent service representative.
3	Defective main board.	Please contact your Agilent service representative.

## **Front Door Error**

#### Error ID: 4350, 4352, 4458

The front door and/or the SLS board are damaged.

Probable cause	Suggested actions
<b>1</b> The sensor on the SLS board is defective.	Please contact your Agilent service representative.
2 The door is bent or the magnet is misplaced/broken.	Please contact your Agilent service representative.

## **Side Door Error**

#### Error ID: 4355, 4459

The side door and/or the main board are damaged.

Probable cause		Suggested actions
1	The door is bent or the magnet is misplaced/broken.	Please contact your Agilent service representative.
2	The sensor on the main board is defective.	Please contact your Agilent service representative.

## **Arm Movement Failed or Arm Movement Timeout**

#### Error ID: 4002

The transport assembly was unable to complete a movement in one of the axes.

The processor defines a certain time window for the successful completion of a movement in any particular axis. The movement and position of the transport assembly is monitored by the encoders on the stepper motors. If the processor does not receive the correct position information from the encoders within the time window, the error message is generated.

#### Axes identification:

- Arm Movement 0 Failed: X-axis.
- Arm Movement 1 Failed: Z-axis.
- Arm Movement 2 Failed: Theta (needle carrier rotation).

Probable cause		Suggested actions
1	Mechanical obstruction.	Ensure unobstructed movement of the transport assembly.
2	High friction in the transport assembly.	Please contact your Agilent service representative.
3	Defective motor assembly.	Please contact your Agilent service representative.
4	Defective sample transport assembly flex board.	Please contact your Agilent service representative.
5	Defective main board.	Please contact your Agilent service representative.

## **Valve to Bypass Failed**

#### Error ID: 4014, 4701

The injection valve failed to switch to the bypass position.

The switching of the injection valve is monitored by two microswitches on the valve assembly. The switches detect the successful completion of the valve movement. If the valve fails to reach the bypass position, or if the microswitch does not close, the error message is generated.

Probable cause	Suggested actions	
<b>1</b> Valve in an intermediate position between the bypass and mainpass positions.	Turn the Autosampler main power OFF and ON.	
<b>2</b> Defective injection valve.	Please contact your Agilent service representative.	
<b>3</b> Defective main board.	Please contact your Agilent service representative.	

## Valve to Mainpass Failed

#### Error ID: 4015

The injection valve failed to switch to the mainpass position.

The switching of the injection valve is monitored by two microswitches on the valve assembly. The switches detect the successful completion of the valve movement. If the valve fails to reach the mainpass position, or if the microswitch does not close, the error message is generated.

P	robable cause	Suggested actions
1	Valve in an intermediate position between the bypass and mainpass positions.	Turn the Autosampler main power OFF and ON.
2	Defective injection valve.	Please contact your Agilent service representative.
3	Defective main board.	Please contact your Agilent service representative.

## **Needle Lock Failed**

#### Error ID: 4702, 4703

The lock assembly on the sampling unit failed to move successfully.

The upper and lower positions of the needle lock are monitored by position sensors on the sampling unit flex board. The sensors detect the successful completion of the needle lock movement. If the needle lock fails to reach the end point, or if the sensors fail to recognize the needle lock movement, the error message is generated.

Probable cause		Suggested actions
1	Defective or dirty position sensor.	Clean the position sensor.
2	Sticking spindle assembly.	Please contact your Agilent service representative.
3	Defective needle drive motor	Please contact your Agilent service representative.
4	Defective main board.	Please contact your Agilent service representative.

## **Needle to Needle Seat Position**

#### Error ID: 4510, 4511, 4714

The needle failed to reach the end position in the needle seat.

The position of the needle is monitored by a position encoder on the needle carrier. If the needle fails to reach the end point, or if the encoder fails to recognize the needle carrier movement, the error message is generated.

Probable cause		Suggested actions
1	Bad sample transport/sampling unit alignment	Do an auto-alignment
2	Bent needle.	Check and exchange the needle assembly if necessary.
3	Missing needle.	Exchange the needle carrier assembly.
4	Blocked seat.	Clean or change the needle seat assembly if necessary.
5	Defective position sensor in the needle carrier assembly.	Please contact your Agilent service representative.
6	Defective main board.	Please contact your Agilent service representative.

## **Needle Carrier Failed**

The needle carrier on the Sample Transport Assembly failed to move correctly.

Probable cause	Suggested actions
1 Defective Z-motor.	Please contact your Agilent service representative.
2 Vial pusher blocked.	Please contact your Agilent service representative.
<b>3</b> Bad needle carrier positioning in X or Theta	Please contact your Agilent service representative.
4 Defective vial pusher sensor.	Please contact your Agilent service representative.
5 Defective main board.	Please contact your Agilent service representative.

## **Missing Vial or Missing Wash Vial**

#### Error ID: 4019, 4034, 4035, 4541, 4542, 4706, 4707

No vial was found in the position defined in the method or sequence.

When the needle carrier moves to a vial and the needle goes into the vial, the position of the needle is monitored by an encoder behind the vial pusher. If no vial is present, the encoder detects an error and the message "missing vial" is generated.

Probable cause		Suggested actions
	<b>1</b> No vial in the position defined in the method or sequence.	Install the sample vial in the correct position, or edit the method or sequence accordingly.
	<b>2</b> Defective needle carrier assembly.	Please contact your Agilent service representative.
	<b>3</b> Defective transport assembly flex board.	Please contact your Agilent service representative.
	4 Defective main board.	Please contact your Agilent service representative.

## **Initialization Failed**

#### Error ID: 4020

The autosampler failed to complete initialization correctly.

The autosampler initialization procedure moves the needle arm and transport assembly to their home positions in a predefined routine. During initialization, the processor monitors the position sensors and motor encoders to check for correct movement. If one or more of the movements is not successful, or is not detected, the error message is generated.

Probable cause		Suggested actions
1	Side door not installed correctly.	<ul> <li>Check if the side door is installed correctly.</li> <li>Check if the magnet is in place in the side door.</li> </ul>
2	Sample transport/sampling unit not aligned correctly.	Do an auto-alignment
3	Mechanical obstruction.	Ensure unobstructed movement of the transport assembly.
4	Defective sampling unit flex board.	Please contact your Agilent service representative.
5	Defective transport assembly flex board.	Please contact your Agilent service representative.
6	Defective sampling unit motor.	Please contact your Agilent service representative.
7	Defective main board.	Please contact your Agilent service representative.

## **Metering Home Failed**

#### Error ID: 4054, 4704

The metering piston has failed to move back to the home position.

The home position sensor on the sampling unit flex board monitors the home position of the piston. If the piston fails to move to the home position, or if the sensor fails to recognize the piston position, the error message is generated.

Probable cause		Suggested actions
1	Dirty or defective sensor.	Please contact your Agilent service representative.
2	Broken plunger.	Exchange the metering plunger and seal.
3	Defective metering-drive motor.	Please contact your Agilent service representative.
4	Defective main board.	Please contact your Agilent service representative.

### **Motor Temperature**

#### Error ID: 4027, 4040, 4261, 4451

One of the motors of the transport assembly has drawn excessive current, causing the motor to become too hot. The processor has switched off the motor to prevent damage to the motor.

Motor identification:

- Motor 0 temperature: X-axis motor.
- Motor 1 temperature: Z-axis motor.
- Motor 2 temperature: Theta motor.

The processor monitors the current drawn by each motor and the time the motor is drawing current. The current drawn by the motors is dependent on the load on each motor (friction, mass of components etc.). If the current drawn is too high, or the time the motor draws current is too long, the error message is generated.

Probable cause		Suggested actions
1 Mec	hanical obstruction.	Ensure unobstructed movement of the transport assembly.
2 High	n friction in the transport assembly.	Please contact your Agilent service representative.
3 Mot	or belt tension too high.	Switch off the module at the power switch. Wait at least 10 minutes before switching on again.
<b>4</b> Defe	ective motor.	Please contact your Agilent service representative.
5 Defe	active transport assembly flex board.	Please contact your Agilent service representative.

## **Invalid Vial Position**

#### **Error ID: 4042**

The vial position defined in the method or sequence does not exist.

The reflection sensors on the transport assembly flex board are used to automatically check which sample trays are installed (coding on tray). If the vial position does not exist in the current sample tray configuration, the error message is generated.

representative.

Probable cause		Suggested actions
1	Incorrect tray installed.	Install the correct trays, or edit the method or sequence accordingly.
2	Incorrect tray definition.	Install the correct trays, or edit the method or sequence accordingly.
3	Incorrect vial positions defined in the method or sequence.	Install the correct trays, or edit the method or sequence accordingly.
4	Tray recognition defective (dirty sample tray or defective transport assembly flex board).	<ul> <li>Ensure the coding surfaces of the sample tray are clean (located at the rear of the sample tray).</li> </ul>
		Please contact your Agilent service

## **Peristaltic Pump Error**

#### Error ID: 4514

The peristaltic pump motor in the autosampler has failed.

The current on the motor is used by the MTP board to monitor the speed of the peristaltic pump motor. If the current falls below a certain value, the error message is generated.

Probable cause	Suggested actions
1 Defective motor.	Please contact your Agilent service representative.
2 Defective SUD board.	Please contact your Agilent service representative.
<b>3</b> Defective main board.	Please contact your Agilent service representative.

## **Vessel or Wash Vessel Error**

#### Error ID: 4540, 4544, 4545, 4705, 4712

The needle does not reach the target position in the vial or in the vessel of the well plate.

The sensor behind the vial pusher in the needle carrier assembly detects the successful completion of the needle movement to the vessel. If the needle fails to reach the end point, the sensor fails to recognize the needle movement and the error message is generated.

Probable cause		Suggested actions
1	Bad vessel definition in the plate configuration.	Check the vessel definition in the plate configuration.
2	Closing mat to rigid/thick.	Check that the closing mat is not too thick.
3	Bad X or Theta positioning.	Please contact your Agilent service representative.
4	Defective encoder on the needle carrier assembly.	Please contact your Agilent service representative.

## **Vessel Stuck to Needle**

#### Error ID: 4453

The vessel sticks to the needle when the needle moves up.

Probable cause	Suggested actions
1 Closing mat to rigid/thick.	Check that the closing mat is not too thick.
<b>2</b> Bad X or Theta positioning and the needle sticks into the wall between two holes.	Please contact your Agilent service representative.
<b>3</b> Defective encoder on the needle carrier assembly.	Please contact your Agilent service representative.

## **Rear Blind Seat Missing**

### Error ID: 4724

Rear blind seat is missing although claimed to exist by main board information – occurs during initialization or if the blind seat location has to be used.

Probable cause	Suggested actions
1 Blind seat is missing.	Install blind seat.

### 8 Error Information

**Module Error Messages** 



# **Test Functions**

9

Introduction 112 System Pressure Test 113 System Pressure Test Evaluation 114 Sample Transport Self Alignment 115 Maintenance Positions 117 Maintenance Positions 117 Change Needle 118 Change Loop Capillary 118 Arm Position 119 Change Needle Carrier 119 Change Metering Device 120 Injector Steps 121 Injector Steps 121 Step Commands 122

This chapter describes the tests for the module.



## Introduction

Interface	Comment	Available Function
Agilent Instrument Utilities	Maintenance tests available	<ul> <li>System Pressure test</li> <li>Sample transport Self Alignment</li> </ul>
Agilent Lab Advisor	All tests are available	<ul> <li>System Pressure test</li> <li>Sample transport Self Alignment</li> </ul>
Agilent ChemStation	No tests available Adding of pressure to chromatographic signals possible	<ul> <li>Pressure</li> <li>Pressure ripple</li> <li>Temperature mainboard</li> </ul>

All tests are described based on the Agilent Lab Advisor Software B.01.04 or above. Other user interfaces may not provide any test or just a few.

For details on the use of the interface refer to the interface documentation.

## **System Pressure Test**

The test determines the leak rate of the system between pump outlet valves and a blank nut. The blank nut can be positioned at different locations in the system before the flow cell, to determine and verify the leak rate of individual modules and components. The test allows for setting the pressure at which the test is performed. The leak rate of high pressure parts are not always a linear function and therefore it is recommended to perform the test at a pressure that correspond to the normal operating pressure of the system.

When In case of a suspected leak. To verify successful execution of maintenance tasks.

Parts required	#	p/n	Description
	1	01080-83202	Blank nut
Preparations	Sol	vents must be pre	esent in both channels.

**1** Run the **System pressure test** with the Agilent Lab Advisor (for further information see Online-Help of user interface).

lest N Aodul	lame	System pressure test for Aladdin G4220A:LP00000003	Description	Preliminary system pressure test f	or Aladdin
	Time	Not defined			
itatus		Passed			
est Pro	cedure		Resu	t	
	_			Name	Value
1.		pump pressure test	Syste	em leak	2.1 bar
2.	Enter th	e test pressure			
3.	Flush th	ne system			
4.	System	checking leak rate of pump			
<b>5</b> .	Insert b	lank nut			
6.	System	checking leak rate of system			
7.	Evaluat	e results			
8.	Restore	system configuration			

Figure 15 System Pressure Test – Result

**System Pressure Test** 

🔆 Pump head	leak test	
⚠	Enter the pressure at which the test will be executed	
	Enter the test pressure	200
		ОК

Figure 16 System Pressure Test – Dynamic pressure input

## **System Pressure Test Evaluation**

#### System Pressure Test Failed

Probable cause	Suggested actions
1 Pump leakages	Perform the Pump Head Leak test.
2 Loose or leaky fittings	Tighten the fittings or replace capillaries.
<b>3</b> Autosampler leakages	Perform the Autosampler Leak test.
4 Thermostatted Column Compartment valve leakages	Replace the TCC valve rotor seal.

### NOTE

- Notice the difference between *error* in the test and a *failed* result! An *error* is caused by an abnormal termination during the operation of the test, whereas a *failed* result indicates that the test results were not within the specified limits.
- Often it is only a damaged blank nut (poorly shaped from over tightening) that causes the test to fail. Before investigating any other possible sources of failure make sure that the blank nut you are using is in a good condition and properly tightened.

## Sample Transport Self Alignment

The sample transport self alignment uses predefined positions on the well plate tray to calibrate the positioning of the needle. The sample transport self alignment is required to compensate for larger deviations in positioning the needle carrier. The sample transport self alignment is required after disassembling the system or when you exchange the sample transport, the sampling unit, the tray or the MTP main board. This function is in the calibration screen of the Lab Advisor.

When After disassembling the module or by larger deviations in the positioning of the needle.

**Preparations** Well plate tray needs to be installed and empty.

### 9 Test Functions

**Sample Transport Self Alignment** 

- lent Lab Advisor General Diagnostic Results Stop Test The sample transport alignment with the sampling unit and the well-plate tray is required to compensate larger deviations in positioning the needle carrier. It is required after disassembling the system or after exchanging ether the sample transport, the sampling unit, the MTP main board or the well plate sampler tray. Description Test Name Transport Alignment 🕘 Print Results Module G4226A:LP00055010 Calibration Descriptions Approx. Time Not defined (2) G1367A/B/C,G1377A,G2258A Status Running G1364A/B/C/D
   ......... Test Procedure 1. Initialize sampler for the calibration 2. Verify that an appropriate well plate tray got installed 3. Ensure that the trav is empty 1. Ensure that the door is closed 5. Align the transport unit and the well plate tray
- **1** Run the **Transport Alignment** with the Agilent Lab Advisor (for further information see Online-Help of user interface).

**Figure 17** Sample Transport Self Alignment– Running

## **Maintenance Positions**

## **Maintenance Positions**

Some maintenance procedures require the needle arm, metering device, 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 Agilent Lab Advisor Software the maintenance positions can be selected from the **Tools** icon.

When

When performing Maintenance on the module.

**1** Run the **Maintenance Positions** with the Agilent Lab Advisor (for further information see Online-Help of user interface).

Agilent Lab Advisor	۲	Agilent Lab A	dvisor	Current User: Current Instrument:	AGILENT\je1208 [Administrator]	
<ul> <li>Lab at a Glance</li> <li>Configuration</li> </ul>		Tools	96.2300		2	
Documentation Firmware Update	- 1	Change Needle	Change Loop Capillary	Arm Position		
Calculators	_	Start	Start	Move Arm	Home	
nstrument	۲	End	End	Park A	um	
System Information Calendar		Change Needle Carrier	Change Metering Device	Status		
<ul> <li>Guided Diagnostics</li> <li>Tests</li> </ul>	- 1	Start	Start	Read	by and a second s	
Calibrations	- 1	End	End	Rese	te	
Early Maintenance Feedback     Status Report						
Logs & Results						
👹 Tools						
Help	۲					
2) 1120 Tools overview						
(2) 1200 Tools overview	_					
					Help	Back

Figure 18 Maintenance Positions– Running

## **Change Needle**

The position is positioning the needle carrier so that there is easy access for changing needle or needle seat. The position is to the far left, and the current to the motors are off, so that the arm can be turned while servicing the module.

Start	

Figure 19 Maintenance Positions– Change Needle

## **Change Loop Capillary**

The **Change Loop Capillary** command positions the arm in the middle of the tray at half height to enable easy exchange of the loop cartridge.

Figure 20 Maintenance Positions– Change Loop Capillary

## **Arm Position**

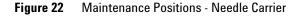
The home position of the autosampler ensures a better access to the tray area and for exchanging trays. When transporting the module it is highly recommended to use the **Park Arm** command, in order to place the Arm in a position for safe transport.

Move Arm Home
Park Arm

Figure 21 Maintenance Positions– Arm Position

## **Change Needle Carrier**

The **Change Needle Carrier** function moves the needle to the front of the autosampler, enabling easy access to the needle carrier mechanism.

- Start moves the needle to the front of the sample-tray area.
- **End** resets the autosampler after the needle carrier has been changed.

## **Change Metering Device**

When removing the metering device is necessary (by exchanging the metering seal for instance), the metering drive needs to be moved to a position at the far back, in order to prevent seal and/or piston damage.

Start	
	_

Figure 23 Maintenance Positions– Change Metering device

## **Injector Steps**

## **Injector Steps**

Each movement of the sampling sequence can be done under manual control. This is useful during troubleshooting, where close observation of each of the sampling steps is required to confirm a specific failure mode or verify successful completion of a repair. Each injector step command actually consists of a series of individual commands that move the autosampler components to predefined positions, enabling the specific step to be done.

#### When

When troubleshooting the module.

**1** Run the **Injector steps** with the Agilent Lab Advisor (for further information see Online-Help of user interface).

Agilent Lab Advisor	۲	Ang P	Agilent La	b Adv	isor	Current User:	AGILENT\je1208 [Administrator]	
🐴 Lab at a Glance		🧼 🗸	ersion B.1.02.	09096.2	300	Current Instrument:	1290 Infinity LC [DE00000000	]
Configuration		Tools						
Documentation								
🏶 Firmware Update					Valve Bypass	# Step Statu:		<u></u>
Calculators	_				Plunger Home			<u></u>
nstrument	۲				Needle Up			5
System Information		Location	P1-A-1		Move To Location			
Calendar		2003001	1.191	-				-
Guided Diagnostics					Needle Into Sample			T <sup>1</sup>
📝 Tests		Volume	1.00	μΙ	Draw			
P Calibrations					Needle Up			
A Early Maintenance Feedback					Needle Into Seat		104000	A High Pe
Status Report							64226	A High Pe Serial 1
Logs & Results					Valve Mainpass			
👹 Tools		Duration	5	s	Needle Up / Valve Mainpass			
telp	۲				Reset			
3 1120 Tools overview					neset			
) 1200 Tools overview								

Figure 24 Injector Steps– Running

## **Step Commands**

Step	Action	Comments
Valve Bypass	Switches injection valve to the bypass position.	
Plunger Home	Moves the plunger to the home position.	
Needle Up	Lifts the needle arm to the upper position.	Command also switches the valve to bypass if it is not already in that position.
Move to Location	Move the needle arm to the vial location on the plate.	
Needle into Sample	Lowers the needle into the vial.	
Draw	Metering device draws the defined injection volume.	Command lifts the needle, and lowers the needle into the sample. Command can be done more than once, maximum draw volume of 20 µL (for 40 µL and 120 µL hardware changes are required see multi-draw) cannot be exeeded. Use <b>Plunger Home</b> to reset the metering device.
Needle Up	Lifts the needle out of the vial.	
Needle into Seat	Lowers the needle arm into the seat.	
Valve Mainpass	Switches the injection valve to the mainpass position.	
Needle Up/Mainpass	Moves needle arm to waste position and switches the injection valve to the mainpass position.	

### Table 6Step Commands



Introduction to Maintenance 124 Warnings and Cautions 125 Overview of Maintenance 127 Cleaning the module 128 Removing the needle assembly 129 Installing the needle assembly 132 Exchanging the Needle Seat 135 Replacing the Rotor seal 138 Removing the metering seal 142 Installing the metering seal 145 **Replacing Peristaltic Pump Cartridge** 147 Installing the Interface Board 150 Replacing the Module Firmware 151

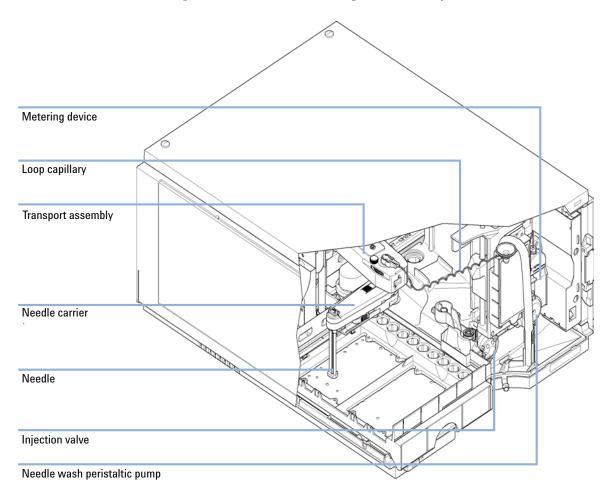
This chapter describes the maintenance of the Autosampler

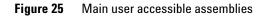


**Introduction to Maintenance** 

## Introduction to Maintenance

Figure 25 on page 124 shows the main user accessible assemblies of the autosampler. These parts can be accessed from the front (simple repairs) and don't require to remove the autosampler from the system stack.





## **Warnings and Cautions**

## WARNING Electrical shock

Repair work at the module can lead to personal injuries, e.g. shock hazard, when the cover is opened.

- → Do not remove the cover of the module.
- → Only certified persons are authorized to carry out repairs inside the module.

### WARNING

#### Personal injury or damage to the product

Agilent is not responsible for any damages caused, in whole or in part, by improper use of the products, unauthorized alterations, adjustments or modifications to the products, failure to comply with procedures in Agilent product user guides, or use of the products in violation of applicable laws, rules or regulations.

Use your Agilent products only in the manner described in the Agilent product user guides.

### WARNING

#### Sharp metal edges

Sharp-edged parts of the equipment may cause injuries.

To prevent personal injury, be careful when getting in contact with sharp metal areas.

Warnings and Cautions

### WARNING Toxic, flammable and hazardous solvents, samples and reagents

#### The handling of solvents, samples and reagents can hold health and safety risks.

- → When working with these substances observe appropriate safety procedures (for example by wearing goggles, safety gloves and protective clothing) as described in the material handling and safety data sheet supplied by the vendor, and follow good laboratory practice.
- The volume of substances should be reduced to the minimum required for the analysis.
- → Do not operate the instrument in an explosive atmosphere.

### CAUTION

Safety standards for external equipment

→ If you connect external equipment to the instrument, make sure that you only use accessory units tested and approved according to the safety standards appropriate for the type of external equipment.

#### CAUTION

#### Sample degradation and contamination of the instrument

Metal parts in the flow path can interact with the bio-molecules in the sample leading to sample degradation and contamination.

- → For bio-inert applications, always use dedicated bio-inert parts, which can be identified by the bio-inert symbol or other markers described in this manual.
- → Do not mix bio-inert and non-inert modules or parts in a bio-inert system.

### NOTE

The electronics of the autosampler will not allow operation of the autosampler when the top cover and the top foam are removed. A safety light switch on the main board will inhibit the operation of the fan immediately. Voltages for the other electronic components will be turned off after 30 seconds. The status lamp will light up red and an error will be logged into the logbook of the user interface. Always operate the autosampler with the top covers in place.

## **Overview of Maintenance**

The following pages describe maintenance (simple repairs) of the autosampler that can be carried out without opening the main cover.

Table 7	Overview of Maintenance
---------	-------------------------

Procedure	Typical Frequency	Notes
Change needle/needle seat	60.000 needle into seat	
Change metering seal	30.000 injections	
Peristaltic pump cartridge	3000 hours on-time	
Change rotor seal	30.000 injections	

### 10 Maintenance Cleaning the module

## **Cleaning the module**

To keep the module case clean, use a soft cloth slightly dampened with water, or a solution of water and mild detergent.

# **WARNING** Liquid dripping into the electronic compartment of your module can cause shock hazard and damage the module

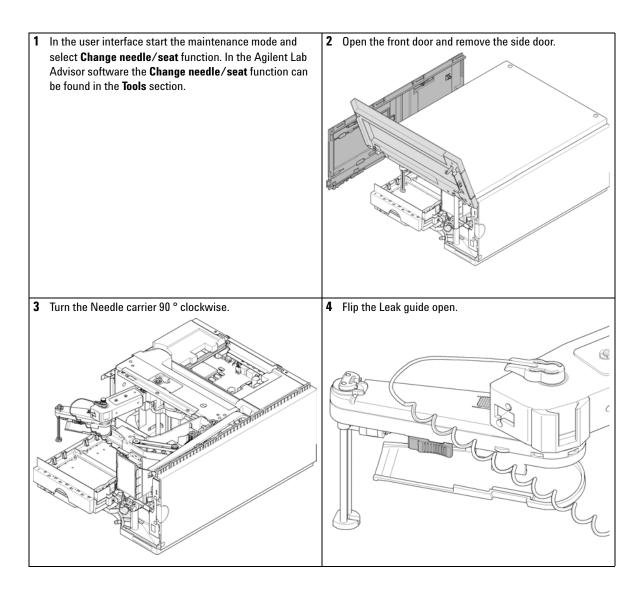
- → Do not use an excessively damp cloth during cleaning.
- → Drain all solvent lines before opening any connections in the flow path.

## Removing the needle assembly

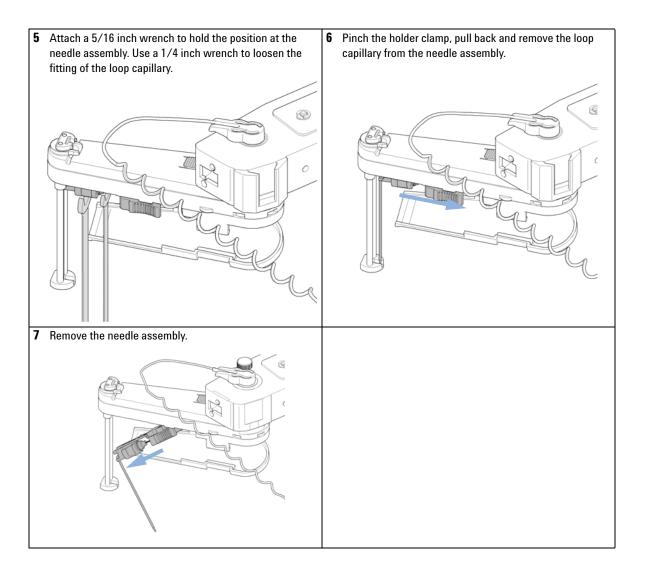
When	When the limit in the needle into seat counter in the EMF is exceeded or when needle shows indications of damage, blockage or leaks.			
Tools required	•	escription 4 inch wrench		
Parts required	<b># p/n</b> 1 G5667-87200	<b>Description</b> Needle assembly (bio-inert)		
Preparations	In order to avoid leaks, close the shutoff valves in the pump or remove tubings from solvent bottles.			
WARNING	<ul> <li>Risk of injury by uncovered needle</li> <li>An uncovered needle is a risk of harm to the operator.</li> <li>→ Be careful working at the needle carrier assembly.</li> <li>→ Use the silicon safety tube supplied with every new needle.</li> </ul>			

**NOTE** It is recommended to always exchange the needle assembly and the needle seat at the same time to prevent premature leakage.

Removing the needle assembly



Removing the needle assembly



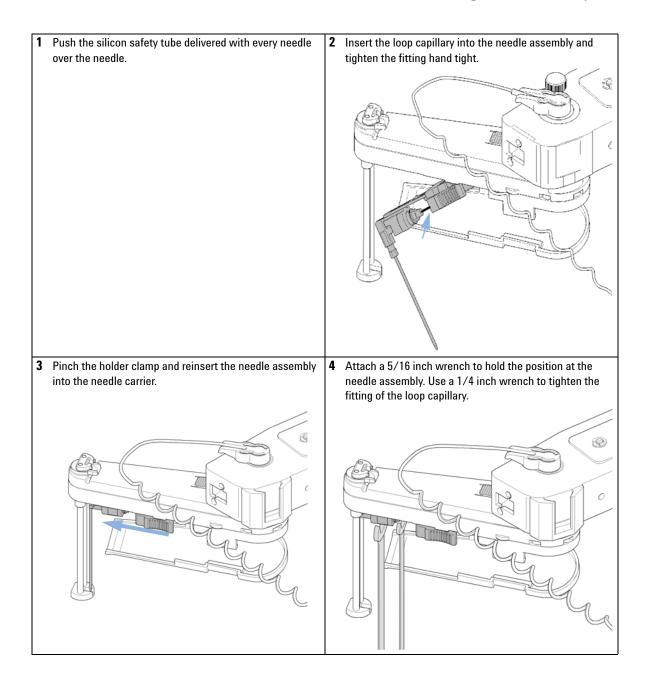
Installing the needle assembly

## Installing the needle assembly

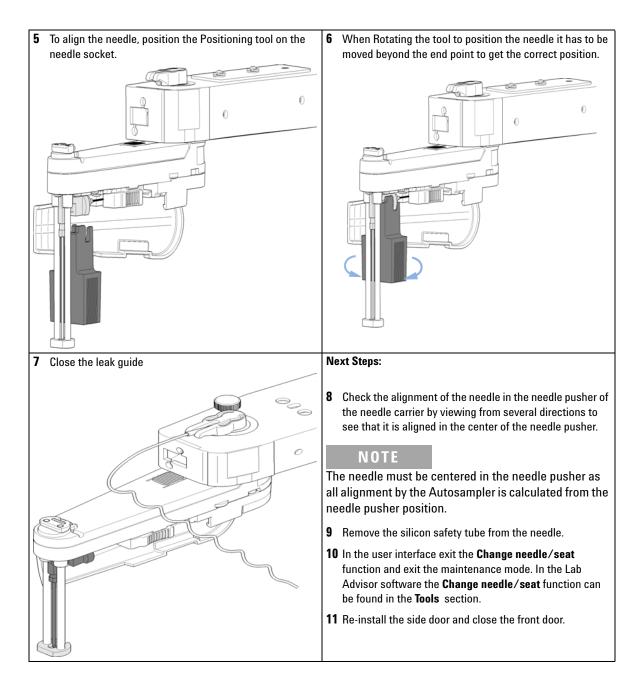
When	When the limit in the needle into seat counter in the EMF is exceeded or when needle shows indications of damage, blockage or leaks.			
Tools required	•	escription 4 inch wrench		
Parts required	# p/n 1 G5667-87200	<b>Description</b> Needle assembly (bio-inert)		
Preparations	In order to avoid leaks, close the shutoff valves in the pump or remove tubings from solvent bottles.			
WARNING	<ul> <li>Risk of injury by uncovered needle</li> <li>An uncovered needle is a risk of harm to the operator.</li> <li>→ Be careful working at the needle carrier assembly.</li> <li>→ Use the silicon safety tube supplied with every new needle.</li> </ul>			

**NOTE** It is recommended to always exchange the needle assembly and the needle seat at the same time to prevent premature leakage.

Installing the needle assembly



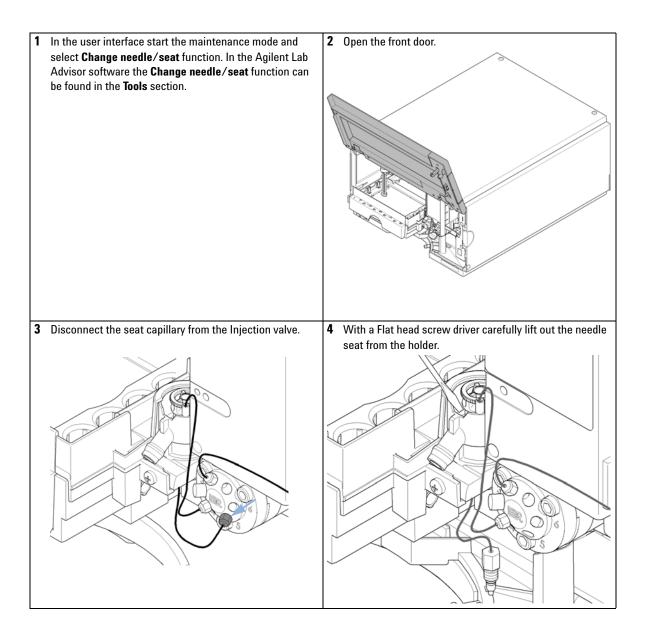
Installing the needle assembly



# **Exchanging the Needle Seat**

When	When seat is visibly damaged, blocked or leaks.				
Tools required	p/n De	escription			
	8710-0510 1/4	inch wrench			
	Fla	at head screwdriver			
Parts required	# p/n	Description			
	1 G5667-87017	Seat assembly (bio-inert), 0.17 mm ID, 100 mm			
Preparations	In order to avoid lea	aks, close the shutoff valves in the pump or remove tubings from solvent bottles.			
WARNING	Risk of injury by	uncovered needle			
	An uncovered needle is a risk of harm to the operator.				
	→ Be careful working at the needle carrier assembly.				
	→ Use the silico	ightarrow Use the silicon safety tube supplied with every new needle.			

**Exchanging the Needle Seat** 



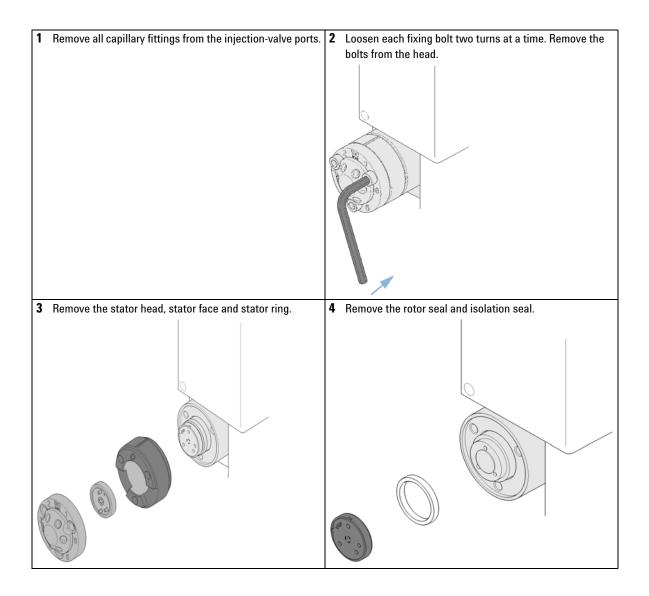
**Exchanging the Needle Seat** 

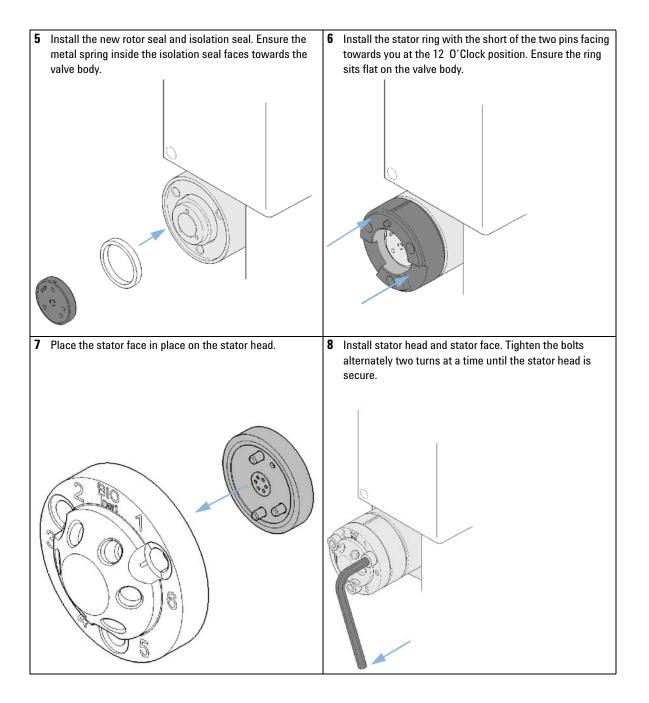
#### Next Steps:

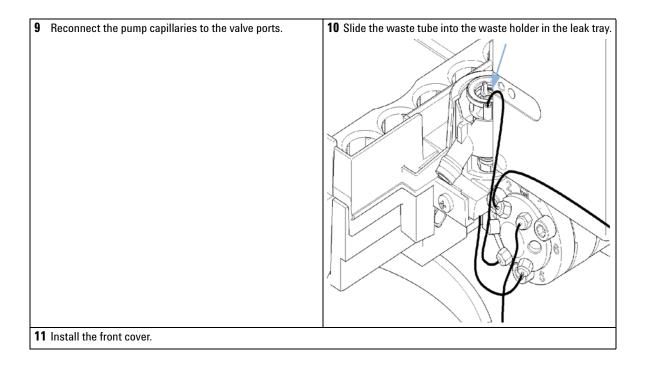
- 5 Insert the new Needle seat. Press it firmly in position.
- 6 Connect capillary to valve.
- 7 In the user interface exit the **Change needle/seat** function and exit the maintenance mode. In the Lab Advisor software the **Change needle/seat** function can be found in the **Tools** section.

Replacing the Rotor seal

When	Poor injection-volume reproducibility			
	Leaking injection	on valve		
Tools required	Description			
	Wrench 1/4 ind	ch		
	Hexagonal key,	9/64 inch		
Parts required	p/n	Description		
	5068-0099	Rotor seal, 3 grooves, max. 600 bar		
Preparations	Remove front c	over.		
	Remove the lea	ık tubing (if necessary).		
CAUTION Damage of the stator face		ne stator face		
		ce is held in place by the stator head. When removing the stator head, the ight fall out of the valve.		
	→ When rem	ightarrow When removing the stator head, ensure the stator face does not fall out of the valve.		





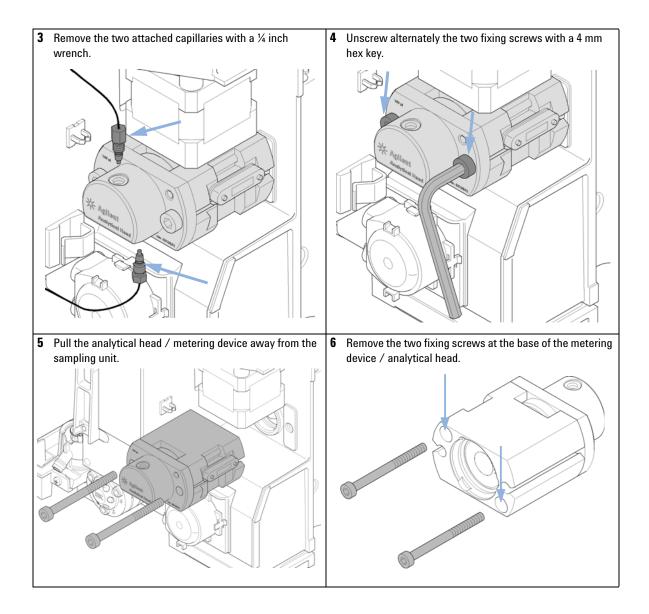


Removing the metering seal

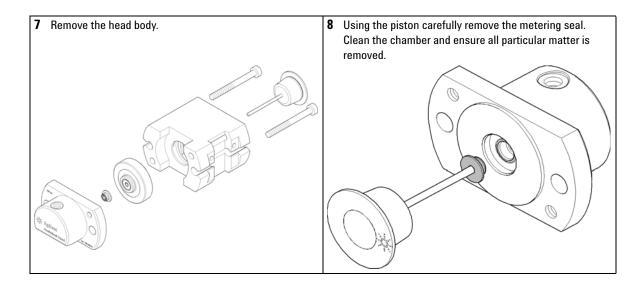
# Removing the metering seal

When	When poor injection volume reproducibility or when metering device $\prime$ analytical head is leaking.			
Tools required Parts required	8710-0510	Description ¼ inch wrench 4 mm Hex key Description 3 Piston seal (bio-inert)		
select <b>Change me</b> Lab Advisor softw	ce start the mainten stering device functi are the Change met und in the Tools sec	ion. In the Agilent tering device		

Removing the metering seal



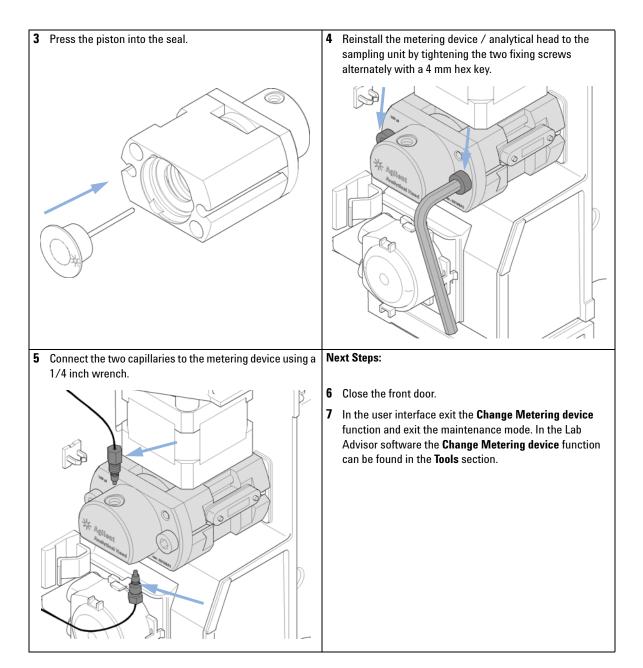
Removing the metering seal



# Installing the metering seal

When	When poor injection volume reproducibility or when metering device $/$ analytical head is leaking.		
Tools required	8710-0510	<b>Description</b> ¼ inch wrench 4 mm Hex key	
Parts required	<b># p/n</b> 1 G5611-21503	<b>Description</b> Piston seal (bio-inert)	
Preparations	Removing the metering seal, see "Removing the metering seal" on page 142.		
	itering seal. Press it f ngle as it might defor		

Installing the metering seal



# **Replacing Peristaltic Pump Cartridge**

When	Tubing blocked or b
------	---------------------

Parts required

NOTE

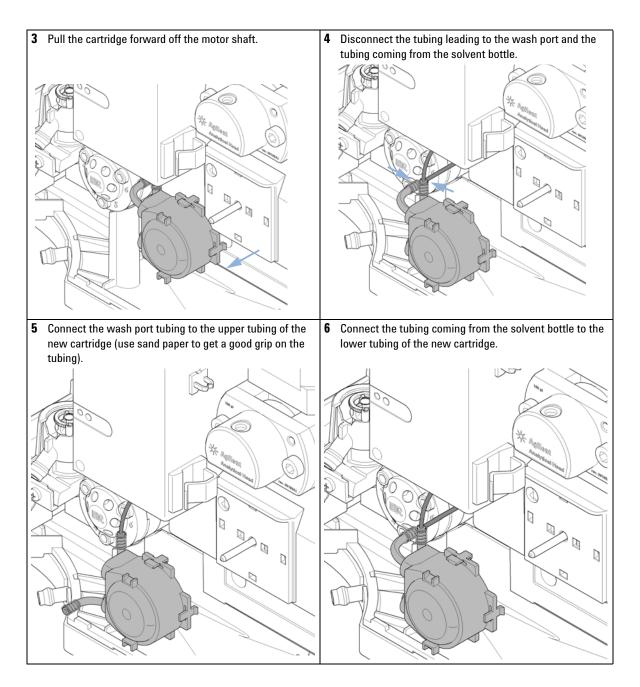
broken

Description # p/n Peristaltic pump cartridge 5065-4445 1

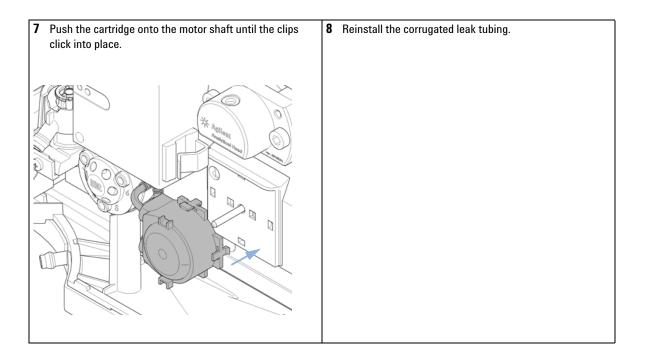
The peristaltic pump cartridge is a replaceable unit. The tubing inside the pump is not replaceable.

<b>1</b> Remove the corrugated leak tubing.	2 Press the two clips on the front of the peristaltic pump cartridge.

**Replacing Peristaltic Pump Cartridge** 



**Replacing Peristaltic Pump Cartridge** 



Installing the Interface Board

# Installing the Interface Board

When	At installation or when defective.		
Tools required	<b>Description</b> Flat head screwdriver		
Parts required	# Description       1 Interface board		
CAUTION	Electronic boards are sensitive to electrostatic discharge (ESD) and should be handled with care so as not to damage them. Touching electronic boards and components can cause electrostatic discharge.		
	ESD can damage electronic boards and components.		
	→ Be sure to hold the board by the edges and do not touch the electrical components. Always use an ESD protection (for example, an ESD wrist strap) when handling electronic boards and components.		
	<b>1</b> Switch OFF the autosampler at the main power switch.		
	<b>2</b> Disconnect cables from the interface board connectors.		
	<ul><li>3 Loosen the screws. Slide out the interface board from the autosampler.</li><li>4 Install the interface board. Secure the screws.</li></ul>		
	<b>5</b> Reconnect the cables to the board connectors.		
Screws			

# **Replacing the Module Firmware**

When	<ul> <li>The installation of newer firmware might be necessary</li> <li>if a newer version solves problems of older versions or</li> <li>to keep all systems on the same (validated) revision.</li> </ul>				
	<ul> <li>The installation of older firmware might be necessary</li> <li>to keep all systems on the same (validated) revision or</li> <li>if a new module with newer firmware is added to a system or</li> <li>if third part control software requires a special version.</li> </ul>				
Tools required	Description				
	LAN/RS-232 Firmware Update Tool				
OR	Agilent Diagnostic Software				
OR	Instant Pilot G4208A				
	(only if supported by module)				
Parts required	# Description				
	1 Firmware, tools and documentation from Agilent web site				
Preparations	Read update documentation provided with the Firmware Update Tool.				
	To upgrade/downgrade the module's firmware carry out the following steps:				
	1 Download the required module firmware, the latest LAN/RS-232 FW Update Tool and the documentation from the Agilent web.				
	<ul> <li>http://www.chem.agilent.com/scripts/cag_firmware.asp.</li> </ul>				
	<b>2</b> For loading the firmware into the module follow the instructions in the documentation.				
	Module Specific Information				
	There is no specific information for this module.				

**Replacing the Module Firmware** 



# Parts for Maintenance

Overview of Maintenance Parts 154 Vial Trays 155 Recommended Plates and Closing Mats 156 Recommended Vial Plates 157 Accessory Kit 158 Injection Valve Assembly 159 Cover Parts 160 Leak System Parts 161

This chapter provides information on parts material required for the module.



### **11** Parts for Maintenance

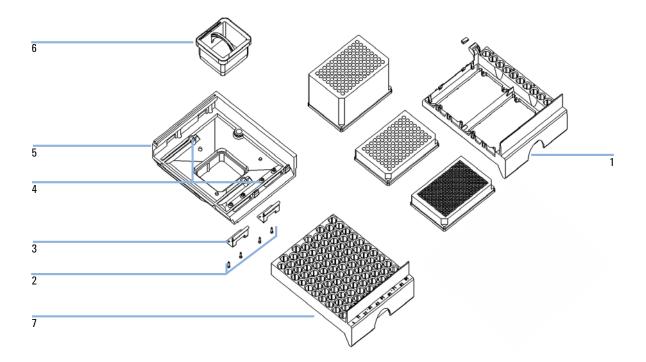
**Overview of Maintenance Parts** 

# **Overview of Maintenance Parts**

BI0 inert For bio-inert modules use bio-inert parts only!

p/n	Description
5068-0099	Rotor seal, 3 grooves, max. 600 bar
G5611-21503	Piston seal (bio-inert)
G5667-87200	Needle assembly (bio-inert)
G5667-87017	Seat assembly (bio-inert), 0.17 mm ID, 100 mm
G5667-60310	Loop capillary (bio-inert)
G5667-60003	Analytical head (bio-inert)
5068-0060	Bio-inert stator head
0100-1851	Stator face, ceramic
G5667-40500	Tool for needle adjustment
5063-6586	Piston
5067-4131	2 position/6port injection valve (bio-inert)

# **Vial Trays**



ltem	p/n	Description
1	G2258-60011	Tray for 2 plates + 10 x 2 mL vials
2	0515-0866	Screws for springs
3	G1313-09101	Spring
4	0570-1574	Spring stud
5	G1329-60000	Tray base
6	G1329-43200	Adapter air channel
	G1367-47200	Plug channel
7	G4226-60021	Tray for 100 micro vials

### **11** Parts for Maintenance

**Recommended Plates and Closing Mats** 

# **Recommended Plates and Closing Mats**

Description (Part Number)	Rows	Columns	Plate height	Volume ( $\mu$ L)	Package
384Agilent (p/n 5042-1388)	16	24	14.4	80	30
384Corning (No Agilent PN)	16	24	14.4	80	
384Nunc (No Agilent PN)	16	24	14.4	80	
96 Agilent (p/n 5042-1386) 96Agilent (p/n 5042-1385)	8	12	14.3	500	10 120
96Agilent conical (p/n 5042-8502)	8	12	17.3	150	25
96CappedAgilent (p/n 5065-4402)	8	12	47.1	300	1
96Corning (No Agilent PN)	8	12	14.3	300	
96CorningV (No Agilent PN)	8	12	14.3	300	
96DeepAgilent31mm (p/n 5042-6454)	8	12	31.5	1000	50
96DeepNunc31mm (No Agilent PN)	8	12	31,5	1000	
96DeepRitter41mm (No Agilent PN)	8	12	41.2	800	
96Greiner (No Agilent PN)	8	12	14.3	300	
96GreinerV (No Agilent PN)	8	12	14.3	250	
96Nunc (No Agilent PN)	8	12	14.3	400	
Closing mat for all 96 Agilent plates (p/n 5042-1389)	8	12			50

#### Table 8 Recommended plates and closing mat

NOTE

Using vessels higher than 41 mm, will result in needle not being able to reach bottom of vessel.

# **Recommended Vial Plates**

p/n	Description
G2255-68700	Vial plate for 54 x 2 mL vials (6/pk)
5022-6539	Vial plate for 15 x 6 mL vials (1/pk)
5022-6538	Vial plate for 27 Eppendorf tubes (1/pk)

#### 11 Parts for Maintenance Accessory Kit

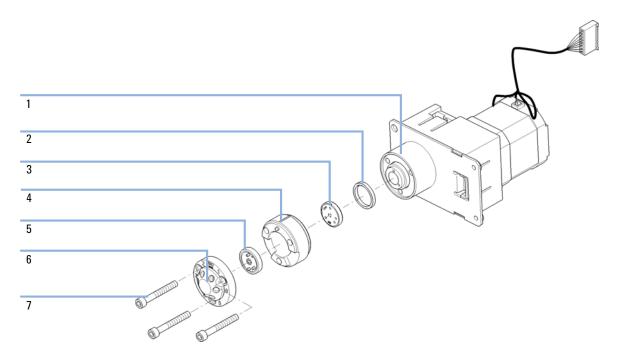
# **Accessory Kit**

p/n	Description
G5667-68755	Accessory kit (bio-inert)
G5667-60500	PEEK/SST capillary (bio-inert) 0.17 mm ID, 400 mm long
5181-1519	CAN cable, Agilent module to module, 1 m
5063-6527	Tubing assembly, i.d. 6 mm, o.d. 9 mm, 1.2 m (to waste)

# **Injection Valve Assembly**

BIU

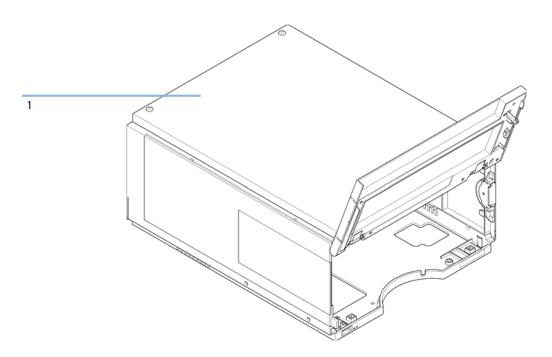
For bio-inert modules use bio-inert parts only!



ltem	p/n	Description
1	5067-4131	2 position/6port injection valve (bio-inert)
2	1535-4045	Isolation seal
3	5068-0099	Rotor seal, 3 grooves, max. 600 bar
4		Rotor head
5	0100-1851	Stator face
6	5068-0060	Bio-inert stator head
7	1535-4857	Stator screws

#### 11 Parts for Maintenance Cover Parts

# **Cover Parts**



ltem	p/n	Description
1	5067-4662	Cabinet kit (base, sides and top)
	5043-0207	Name plate 1260
	G4226-67001	Door repair kit, includes the front door

# Leak System Parts

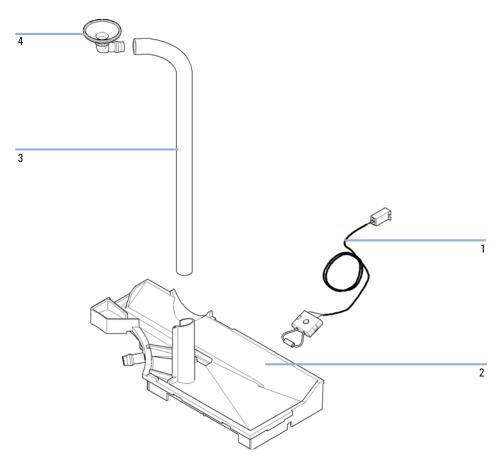
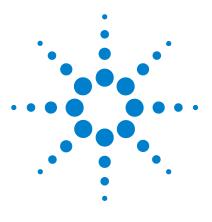


Figure 26 Leak system parts

ltem	p/n	Description
1	5061-3356	Leak sensor
2	G4226-44511	Leak plane
3	0890-1711	Leak tubing 185 mm
4	5041-8388	Leak funnel

## **11** Parts for Maintenance

Leak System Parts



# 12 Identifying Cables

Cable Overview 164 Analog Cables 166 Remote Cables 168 BCD Cables 171 CAN/LAN Cables 173 External Contact Cable 174 Agilent Module to PC 175 Agilent 1200 Module to Printer 176

This chapter provides information on cables used with the 1260 series of HPLC modules.





# **Cable Overview**

# NOTE

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

#### **Analog cables**

p/n	Description
35900-60750	Agilent module to 3394/6 integrators
35900-60750	Agilent 35900A A/D converter
01046-60105	Analog cable (BNC to general purpose, spade lugs)

#### **Remote cables**

p/n	Description
03394-60600	Agilent module to 3396A Series I integrators
	3396 Series II / 3395A integrator, see details in section "Remote Cables" on page 168 $$
03396-61010	Agilent module to 3396 Series III / 3395B integrators
5061-3378	Remote Cable
01046-60201	Agilent module to general purpose

#### **BCD** cables

p/n	Description
03396-60560	Agilent module to 3396 integrators
G1351-81600	Agilent module to general purpose

### **CAN** cables

p/n	Description
5181-1516	CAN cable, Agilent module to module, 0.5 m
5181-1519	CAN cable, Agilent module to module, 1 m

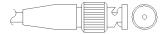
### LAN cables

p/n	Description
5023-0203	Cross-over network cable, shielded, 3 m (for point to point connection)
5023-0202	Twisted pair network cable, shielded, 7 m (for point to point connection)

#### **RS-232** cables

p/n	Description
G1530-60600	RS-232 cable, 2 m
RS232-61600	RS-232 cable, 2.5 m Instrument to PC, 9-to-9 pin (female). This cable has special pin-out, and is not compatible with connecting printers and plotters. It's also called "Null Modem Cable" with full handshaking where the wiring is made between pins 1-1, 2-3, 3-2, 4-6, 5-5, 6-4, 7-8, 8-7, 9-9.
5181-1561	RS-232 cable, 8 m

# **Analog Cables**



One end of these cables provides a BNC connector to be connected to Agilent modules. The other end depends on the instrument to which connection is being made.

## Agilent Module to 3394/6 Integrators

	module	
1		Not connected
2	Shield	Analog -
3	Center	Analog +

# Agilent Module to BNC Connector

p/n 8120-1840	Pin BNC	Pin Agilent module	Signal Name	
x IIIO	Shield	Shield	Analog -	
	Center	Center	Analog +	

# **Agilent Module to General Purpose**

p/n 01046-60105	Pin	Pin Agilent module	Signal Name
	1		Not connected
	2	Black	Analog -
	3	Red	Analog +
	~~ 		

# **Remote Cables**



One end of these cables provides a Agilent Technologies APG (Analytical Products Group) remote connector to be connected to Agilent modules. The other end depends on the instrument to be connected to.

### **Agilent Module to 3396A Integrators**

p/n 03394-60600	Pin 3396A	Pin Agilent module	Signal Name	Active (TTL)
	9	1 - White	Digital ground	
80 15	NC	2 - Brown	Prepare run	Low
	3	3 - Gray	Start	Low
	NC	4 - Blue	Shut down	Low
	NC	5 - Pink	Not connected	
	NC	6 - Yellow	Power on	High
	5,14	7 - Red	Ready	High
	1	8 - Green	Stop	Low
	NC	9 - Black	Start request	Low
	13, 15		Not connected	

### Agilent Module to 3396 Series II / 3395A Integrators

Use the cable Agilent module to 3396A Series I integrators (p/n 03394-60600) and cut pin #5 on the integrator side. Otherwise the integrator prints START; not ready.

p/n 03396-61010	Pin 33XX	Pin Agilent module	Signal Name	Active (TTL)
	9	1 - White	Digital ground	
80 15	NC	2 - Brown	Prepare run	Low
	3	3 - Gray	Start	Low
	NC	4 - Blue	Shut down	Low
	NC	5 - Pink	Not connected	
	NC	6 - Yellow	Power on	High
	14	7 - Red	Ready	High
	4	8 - Green	Stop	Low
	NC	9 - Black	Start request	Low
	13, 15		Not connected	

## Agilent Module to 3396 Series III / 3395B Integrators

## Agilent Module to Agilent 35900 A/D Converters

p/n 5061-3378	Pin 35900 A/D	Pin Agilent module	Signal Name	Active (TTL)
	1 - White	1 - White	Digital ground	
	2 - Brown	2 - Brown	Prepare run	Low
	3 - Gray	3 - Gray	Start	Low
	4 - Blue	4 - Blue	Shut down	Low
	5 - Pink	5 - Pink	Not connected	
$\bigcirc$	6 - Yellow	6 - Yellow	Power on	High
	7 - Red	7 - Red	Ready	High
	8 - Green	8 - Green	Stop	Low
	9 - Black	9 - Black	Start request	Low

p/n 01046-60201	Wire Color	Pin Agilent module	Signal Name	Active (TTL)
	White	1	Digital ground	
	Brown	2	Prepare run	Low
	Gray	3	Start	Low
	Blue	4	Shut down	Low
	Pink	5	Not connected	
	Yellow	6	Power on	High
	Red	7	Ready	High
	Green	8	Stop	Low
	Black	9	Start request	Low

# **Agilent Module to General Purpose**

# **BCD Cables**



One end of these cables provides a 15-pin BCD connector to be connected to the Agilent modules. The other end depends on the instrument to be connected to

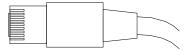
### **Agilent Module to General Purpose**

p/n G1351-81600	Wire Color	Pin Agilent module	Signal Name	BCD Digit
	Green	1	BCD 5	20
I.M.	Violet	2	BCD 7	80
	Blue	3	BCD 6	40
	Yellow	4	BCD 4	10
	Black	5	BCD 0	1
	Orange	6	BCD 3	8
	Red	7	BCD 2	4
	Brown	8	BCD 1	2
	Gray	9	Digital ground	Gray
	Gray/pink	10	BCD 11	800
	Red/blue	11	BCD 10	400
	White/green	12	BCD 9	200
	Brown/green	13	BCD 8	100
	not connected	14		
	not connected	15	+ 5 V	Low

# Agilent Module to 3396 Integrators

p/n 03396-60560	Pin 3396	Pin Agilent module	Signal Name	BCD Digit
	1	1	BCD 5	20
	2	2	BCD 7	80
	3	3	BCD 6	40
	4	4	BCD 4	10
	5	5	BCD0	1
	6	6	BCD 3	8
	7	7	BCD 2	4
	8	8	BCD 1	2
	9	9	Digital ground	
	NC	15	+ 5 V	Low

# **CAN/LAN Cables**



Both ends of this cable provide a modular plug to be connected to Agilent modules CAN or LAN connectors.

#### **CAN Cables**

p/n	Description
5181-1516	CAN cable, Agilent module to module, 0.5 m
5181-1519	CAN cable, Agilent module to module, 1 m

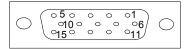
### LAN Cables

p/n	Description
5023-0203	Cross-over network cable, shielded, 3 m (for point to point connection)
5023-0202	Twisted pair network cable, shielded, 7 m (for point to point connection)

**12** Identifying Cables

**External Contact Cable** 

# **External Contact Cable**



One end of this cable provides a 15-pin plug to be connected to Agilent modules interface board. The other end is for general purpose.

### Agilent Module Interface Board to general purposes

p/n G1103-61611	Color	Pin Agilent module	Signal Name
	White	1	EXT 1
	Brown	2	EXT 1
	Green	3	EXT 2
	Yellow	4	EXT 2
	Grey	5	EXT 3
	Pink	6	EXT 3
	Blue	7	EXT 4
	Red	8	EXT 4
	Black	9	Not connected
	Violet	10	Not connected
	Grey/pink	11	Not connected
	Red/blue	12	Not connected
	White/green	13	Not connected
	Brown/green	14	Not connected
	White/yellow	15	Not connected

# **Agilent Module to PC**

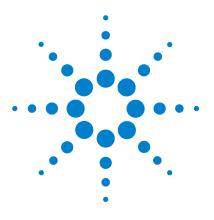
p/n	Description
G1530-60600	RS-232 cable, 2 m
RS232-61600	RS-232 cable, 2.5 m Instrument to PC, 9-to-9 pin (female). This cable has special pin-out, and is not compatible with connecting printers and plotters. It's also called "Null Modem Cable" with full handshaking where the wiring is made between pins 1-1, 2-3, 3-2, 4-6, 5-5, 6-4, 7-8, 8-7, 9-9.
5181-1561	RS-232 cable, 8 m

## **12** Identifying Cables

Agilent 1200 Module to Printer

# **Agilent 1200 Module to Printer**

p/n	Description
5181-1529	Cable Printer Serial & Parallel, is a SUB-D 9 pin female vs. Centronics connector on the other end (NOT FOR FW UPDATE). For use with G1323 Control Module.



# 13 Hardware Information

Firmware Description 178 Boot-up and Initialization Process 181 Interfaces 182 Overview Interfaces 184 Setting the 8-bit Configuration Switch 188 Communication Settings for RS-232C 189 Special Settings 191 Electrical Connections 192 Rear view of the module 193 Information on Instrument Serial Number 193

This chapter describes the autosampler in more detail on hardware and electronics.



# **Firmware Description**

The firmware of the instrument consists of two independent sections:

- a non-instrument specific section, called *resident system*
- an instrument specific section, called *main system*

#### **Resident System**

This resident section of the firmware is identical for all Agilent 1100/1200/1220/1260/1290 series modules. Its properties are:

- the complete communication capabilities (CAN, LAN and RS-232C)
- memory management
- · ability to update the firmware of the 'main system'

#### **Main System**

Its properties are:

- the complete communication capabilities (CAN, LAN and RS-232C)
- memory management
- · ability to update the firmware of the 'resident system'

In addition the main system comprises the instrument functions that are divided into common functions like

- run synchronization through APG remote,
- error handling,
- diagnostic functions,
- or module specific functions like
  - internal events such as lamp control, filter movements,
  - raw data collection and conversion to absorbance.

### **Firmware Updates**

Firmware updates can be done using your user interface:

• PC and Firmware Update Tool with local files on the hard disk

- Instant Pilot (G4208A) with files from a USB Flash Disk
- Agilent Lab Advisor software B.01.03 and above

The file naming conventions are:

PPPP\_RVVV\_XXX.dlb, where

PPPP is the product number, for example, 1315AB for the G1315A/B DAD,

R the firmware revision, for example, A for G1315B or B for the G1315C DAD,

VVV is the revision number, for example 102 is revision 1.02,

XXX is the build number of the firmware.

For instructions on firmware updates refer to section *Replacing Firmware* in chapter "Maintenance" or use the documentation provided with the *Firmware Update Tools*.

Update of main system can be done in the resident system only. Update of the resident system can be done in the main system only.

Main and resident firmware must be from the same set.

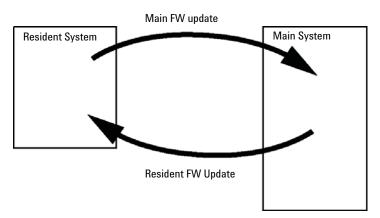


Figure 27 Firmware Update Mechanism

NOTE

#### **13** Hardware Information

**Firmware Description** 

### NOTE

Some modules are limited in downgrading due to their main board version or their initial firmware revision. For example, a G1315C DAD SL cannot be downgraded below firmware revision B.01.02 or to a A.xx.xx.

Some modules can be re-branded (e.g. G1314C to G1314B) to allow operation in specific control software environments. In this case the feature set of the target type are use and the feature set of the original are lost. After re-branding (e.g. from G1314B to G1314C), the original feature set is available again.

All these specific informations are described in the documentation provided with the firmware update tools.

The firmware update tools, firmware and documentation are available from the Agilent web.

 http://www.chem.agilent.com/EN-US/SUPPORT/DOWNLOADS/FIRMWARE/ Pages/LC.aspx

# **Boot-up and Initialization Process**

#### CAUTION

#### Obstruction of transport unit

Any obstruction of the transport unit during the initialization process will result in a wrong transmission ratio and thus wrong needle positions.

- → Make sure no vials or other material gets into the X-slide.
- 1 Firmware Boot Process.
  - a Start Boot Loader.
  - **b** Boot main firmware.

OR

Boot resident firmware (if set in VRAM, by DIP switch or if no/wrong main FW is found).

- 2 Initialize Transport Unit.
  - a Switch injection valve to bypass position.
  - **b** Find initial positions for X,Z and theta motors.
  - c Check belt tension of theta motor.
  - **d** Determine transmission ratio for X and theta axes.
    - Turn needle carrier fully counter-clockwise (= theta min).
    - Move X-slide into left end-stop (= X min).
    - Move X-slide into right end-stop (= X max).
    - Rotate needle carrier fully clockwise (= theta max, happens at the same time as step iii.).
- **3** Read RFID tag of Sampling Unit.
- 4 Read RFID tag of sample tray (if tray is different from last time).
- **5** Move needle into needle seat to determine the seat depth.
- **6** Move needle into seat (use depth value from step 5).
- **7** Lower the needle lock.
- 8 Switch the injection valve to mainpass.

## **13** Hardware Information Interfaces

# Interfaces

The Agilent 1200 Infinity Series modules provide the following interfaces:

Table 9	Agilent 1200 Infinity Series Interfaces	
---------	---	--

Module	CAN	LAN/BCD (optional)	LAN (on-board)	RS-232	Analog	APG Remote	Special
Pumps							
G1310B Iso Pump G1311B Quat Pump G1311C Quat Pump VL G1312B Bin Pump G1312C Bin Pump VL 1376A Cap Pump G2226A Nano Pump G5611A Bio-inert Quat Pump	2	Yes	No	Yes	1	Yes	
G4220A/B Bin Pump	2	No	Yes	Yes	No	Yes	
G1361A Prep Pump	2	Yes	No	Yes	No	Yes	CAN-DC- OUT for CAN slaves
Samplers							
G1329B ALS G2260A Prep ALS	2	Yes	No	Yes	No	Yes	THERMOSTAT for G1330B
G1364B FC-PS G1364C FC-AS G1364D FC-μS G1367E HiP ALS G1377A HiP micro ALS G2258A DL ALS G5664A Bio-inert FC-AS G5667A Bio-inert Autosampler	2	Yes	No	Yes	No	Yes	THERMOSTAT for G1330B CAN-DC- OUT for CAN slaves
G4226A ALS	2	Yes	No	Yes	No	Yes	
Detectors							
G1314B VWD VL G1314C VWD VL+	2	Yes	No	Yes	1	Yes	

Module	CAN	LAN/BCD (optional)	LAN (on-board)	RS-232	Analog	APG Remote	Special
G1314E/F VWD	2	No	Yes	Yes	1	Yes	
G4212A/B DAD	2	No	Yes	Yes	1	Yes	
G1315C DAD VL+ G1365C MWD G1315D DAD VL G1365D MWD VL	2	No	Yes	Yes	2	Yes	
G1321B FLD G1362A RID	2	Yes	No	Yes	1	Yes	
G4280A ELSD	No	No	No	Yes	Yes	Yes	EXT Contact AUTOZERO
Others							
G1170A Valve Drive	2	No	No	No	No	No	Requires a HOST module with on-board LAN (e.g. G4212A or G4220A with minimum firmware B.06.40 or C.06.40) or with additional G1369C LAN Card
G1316A/C TCC	2	No	No	Yes	No	Yes	
G1322A DEG	No	No	No	No	No	Yes	AUX
G1379B DEG	No	No	No	Yes	No	Yes	
G4225A DEG	No	No	No	Yes	No	Yes	
G4227A Flex Cube	2	No	No	No	No	No	
G4240A CHIP CUBE	2	Yes	No	Yes	No	Yes	CAN-DC- OUT for CAN slaves THERMOSTAT for G1330A/B (NOT USED)

## Table 9 Agilent 1200 Infinity Series Interfaces

# NOTE

The detector (DAD/MWD/FLD/VWD/RID) is the preferred access point for control via LAN. The inter-module communication is done via CAN.

# **13** Hardware Information

Interfaces

- CAN connectors as interface to other modules
- LAN connector as interface to the control software
- RS-232C as interface to a computer
- · REMOTE connector as interface to other Agilent products
- Analog output connector(s) for signal output

# **Overview Interfaces**

#### CAN

The CAN is inter-module communication interface. It is a 2-wire serial bus system supporting high speed data communication and real-time requirement.

#### LAN

The modules have either an interface slot for an LAN card (e.g. Agilent G1369B/C LAN Interface) or they have an on-board LAN interface (e.g. detectors G1315C/D DAD and G1365C/D MWD). This interface allows the control of the module/system via a PC with the appropriate control software.

#### NOTE

If an Agilent detector (DAD/MWD/FLD/VWD/RID) is in the system, the LAN should be connected to the DAD/MWD/FLD/VWD/RID (due to higher data load). If no Agilent detector is part of the system, the LAN interface should be installed in the pump or autosampler.

#### **RS-232C** (Serial)

The RS-232C connector is used to control the module from a computer through RS-232C connection, using the appropriate software. This connector can be configured with the configuration switch module at the rear of the module. Refer to *Communication Settings for RS-232C*.

# NOTE

There is no configuration possible on main boards with on-board LAN. These are pre-configured for

- 19200 baud,
- 8 data bit with no parity and
- one start bit and one stop bit are always used (not selectable).

The RS-232C is designed as DCE (data communication equipment) with a 9-pin male SUB-D type connector. The pins are defined as:

Pin	Direction	Function
1	In	DCD
2	In	RxD
3	Out	TxD
4	Out	DTR
5		Ground
6	In	DSR
7	Out	RTS
8	In	CTS
9	In	RI

 Table 10
 RS-232C Connection Table

#### **13** Hardware Information

Interfaces

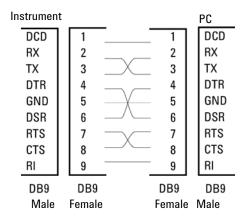


Figure 28 RS-232 Cable

## **Analog Signal Output**

The analog signal output can be distributed to a recording device. For details refer to the description of the module's main board.

#### **APG Remote**

The APG Remote connector may be used in combination with other analytical instruments from Agilent Technologies if you want to use features as common shut down, prepare, and so on.

Remote control allows easy connection between single instruments or systems to ensure coordinated analysis with simple coupling requirements.

The subminiature D connector is used. The module provides one remote connector which is inputs/outputs (wired- or technique).

To provide maximum safety within a distributed analysis system, one line is dedicated to **SHUT DOWN** the system's critical parts in case any module detects a serious problem. To detect whether all participating modules are switched on or properly powered, one line is defined to summarize the **POWER ON** state of all connected modules. Control of analysis is maintained by signal readiness **READY** for next analysis, followed by **START** of run and optional **STOP** of run triggered on the respective lines. In addition **PREPARE** and **START REQUEST** may be issued. The signal levels are defined as:

- standard TTL levels (0 V is logic true, + 5.0 V is false),
- fan-out is 10,

- input load is 2.2 kOhm against + 5.0 V, and
- output are open collector type, inputs/outputs (wired- or technique).

All common TTL circuits operate with a 5 V power supply. A TTL signal is defined as "low" or L when between 0 V and 0.8 V and "high" or H when between 2.0 V and 5.0 V (with respect to the ground terminal).

Pin	Signal	Description
1	DGND	Digital ground
2	PREPARE	(L) Request to prepare for analysis (for example, calibration, detector lamp on). Receiver is any module performing pre-analysis activities.
3	START	(L) Request to start run / timetable. Receiver is any module performing run-time controlled activities.
4	SHUT DOWN	(L) System has serious problem (for example, leak: stops pump). Receiver is any module capable to reduce safety risk.
5		Not used
6	POWER ON	(H) All modules connected to system are switched on. Receiver is any module relying on operation of others.
7	READY	(H) System is ready for next analysis. Receiver is any sequence controller.
8	STOP	(L) Request to reach system ready state as soon as possible (for example, stop run, abort or finish and stop injection). Receiver is any module performing run-time controlled activities.
9	START REQUEST	(L) Request to start injection cycle (for example, by start key on any module). Receiver is the autosampler.

 Table 11
 Remote Signal Distribution

NOTE

# **Special Interfaces**

Some modules have module specific interfaces/connectors. They are described in the module documentation.

# Setting the 8-bit Configuration Switch

The 8-bit configuration switch is located at the rear of the module.

This module does not have its own on-board LAN interface. It can be controlled through the LAN interface of another module, and a CAN connection to that module.

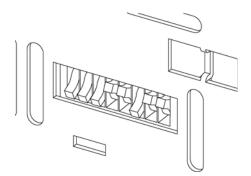


Figure 29 Configuration switch (settings depend on configured mode)

All modules without on-board LAN:

- default should be ALL DIPS DOWN (= best settings)
  - Bootp mode for LAN and
  - \* 19200 baud, 8 data bit / 1 stop bit with no parity for RS-232
- DIP 1 DOWN and DIP 2 UP allows special RS-232 settings
- for boot/test modes DIPS 1+2 must be UP plus required mode

#### NOTE

For normal operation use the default (best) settings.

Switch settings provide configuration parameters for serial communication protocol and instrument specific initialization procedures.

NOTE

With the introduction of the Agilent 1260 Infinity, all GPIB interfaces have been removed. The preferred communication is LAN.

Setting the 8-bit Configuration Switch

# **NOTE** The following tables represent the configuration switch settings for the modules without on-board LAN only.

Mode Select	1	2	3	4	5	6	7	8
RS-232C	0	1	Baudrate			Data Bits	Parity	
Reserved	1	0		Reserved				
TEST/BOOT	1	1	RSVD SYS RSV				RSVD	FC

#### Table 12 8-bit Configuration Switch (without on-board LAN)

NOTE

The LAN settings are done on the LAN Interface Card G1369B/C. Refer to the documentation provided with the card.

# **Communication Settings for RS-232C**

The communication protocol used in the column compartment supports only hardware handshake (CTS/RTR).

Switches 1 in down and 2 in up position define that the RS-232C parameters will be changed. Once the change has been completed, the column instrument must be powered up again in order to store the values in the non-volatile memory.

	Mode Select	1	2	3	4	5	6	7	8
ĺ	RS-232C	0	1		Baudrate		Data Bits	Pari	ty

Table 13 Communication Settings for RS-232C Communication (without on-board LAN)

Use the following tables for selecting the setting which you want to use for RS-232C communication. The number 0 means that the switch is down and 1 means that the switch is up.

## **13** Hardware Information

Setting the 8-bit Configuration Switch

Switches		Baud Rate		Switches	Baud Rate		
3	4	5		3	4	5	
0	0	0	9600	1	0	0	9600
0	0	1	1200	1	0	1	14400
0	1	0	2400	1	1	0	19200
0	1	1	4800	1	1	1	38400

**Table 14** Baudrate Settings (without on-board LAN)

 Table 15
 Data Bit Settings (without on-board LAN)

Switch 6	Data Word Size
0	7 Bit Communication
1	8 Bit Communication

 Table 16
 Parity Settings (without on-board LAN)

Swite	ches	Parity
7	8	
0	0	No Parity
0	1	Odd Parity
1	1	Even Parity

One start bit and one stop bit are always used (not selectable).

Per default, the module will turn into 19200 baud, 8 data bit with no parity.

# **Special Settings**

The special settings are required for specific actions (normally in a service case).

## **Boot-Resident**

Firmware update procedures may require this mode in case of firmware loading errors (main firmware part).

If you use the following switch settings and power the instrument up again, the instrument firmware stays in the resident mode. It is not operable as a module. It only uses basic functions of the operating system for example, for communication. In this mode the main firmware can be loaded (using update utilities).

 Table 17
 Boot Resident Settings (without on-board LAN)

Mode Select	SW1	SW2	SW3	SW4	SW5	SW6	SW7	SW8
TEST/BOOT	1	1	0	0	1	0	0	0

## **Forced Cold Start**

A forced cold start can be used to bring the module into a defined mode with default parameter settings.

CAUTION
 Loss of data
 Forced cold start erases all methods and data stored in the non-volatile memory.
 Exceptions are calibration settings, diagnosis and repair log books which will not be erased.
 → Save your methods and data before executing a forced cold start.

If you use the following switch settings and power the instrument up again, a forced cold start has been completed.

 Table 18
 Forced Cold Start Settings (without on-board LAN)

Mode Select	SW1	SW2	SW3	SW4	SW5	SW6	SW7	SW8
TEST/BOOT	1	1	0	0	1	0	0	1

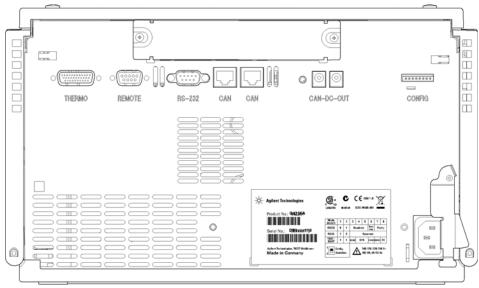
#### 13 Hardware Information Electrical Connections

# **Electrical Connections**

- The CAN bus is a serial bus with high speed data transfer. The two connectors for the CAN bus are used for internal module data transfer and synchronization.
- One analog output provides signals for integrators or data handling systems.
- The REMOTE connector may be used in combination with other analytical instruments from Agilent Technologies if you want to use features such as start, stop, common shut down, prepare, and so on.
- With the appropriate software, the RS-232C connector may be used to control the module from a computer through a RS-232C connection. This connector is activated and can be configured with the configuration switch.
- The power input socket accepts a line voltage of 100 240 VAC  $\pm 10$  % with a line frequency of 50 or 60 Hz. Maximum power consumption varies by module. There is no voltage selector on your module because the power supply has wide-ranging capability. There are no externally accessible fuses, because automatic electronic fuses are implemented in the power supply.

# NOTE

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



# Rear view of the module

Figure 30 Rear view of the module

# **Information on Instrument Serial Number**

The serial number information on the instrument labels provide the following information:

CCXZZ00000	Format
CC	Country of manufacturing • DE = Germany • JP = Japan • CN = China
Х	Alphabetic character A-Z (used by manufacturing)
ZZ	Alpha-numeric code 0-9, A-Z, where each combination unambiguously denotes a module (there can be more than one code for the same module)
00000	Serial number

#### **13** Hardware Information

**Electrical Connections** 



# 14 Appendix

General Safety Information 196 Lithium Batteries Information 199 The Waste Electrical and Electronic Equipment (WEEE) Directive (2002/96/EC) 200 Radio Interference 201 Sound Emission 202 Solvent Information 203 Agilent Technologies on Internet 205

This chapter provides addition information on safety, legal and web.



## 14 Appendix

**General Safety Information** 

# **General 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.

## WARNING

#### Ensure the proper usage of the equipment.

The protection provided by the equipment may be impaired.

The operator of this instrument is advised to use the equipment in a manner as specified in this manual.

# **Safety Standards**

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

# 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 fuse holders must be avoided.

Some adjustments described in the manual, are made with power supplied to the instrument, and protective covers removed. Energy available at many points may, if contacted, result in personal injury.

Any adjustment, maintenance, and repair of the opened instrument under voltage should be avoided whenever possible. When inevitable, this has to be carried out by a skilled person who is aware of the hazard involved. Do not attempt internal service or adjustment unless another person, capable of rendering first aid and resuscitation, is present. Do not replace components with power cable connected.

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.

When working with solvents, observe appropriate safety procedures (for example, goggles, safety gloves and protective clothing) as described in the material handling and safety data sheet by the solvent vendor, especially when toxic or hazardous solvents are used.

#### **14** Appendix

**General Safety Information** 

# **Safety Symbols**

Symbol	Description
$\wedge$	The apparatus is marked with this symbol when the user should refer to the instruction manual in order to protect risk of harm to the operator and to protect the apparatus against damage.
4	Indicates dangerous voltages.
	Indicates a protected ground terminal.
	Indicates eye damage may result from directly viewing the light produced by the deuterium lamp used in this product.
<u>A</u>	The apparatus is marked with this symbol when hot surfaces are available and the user should not touch it when heated up.

#### Table 19Safety Symbols

# WARNING

#### **A WARNING**

#### alerts you to situations that could cause physical injury or death.

→ 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 loss of data, or damage of equipment.

→ Do not proceed beyond a caution until you have fully understood and met the indicated conditions.

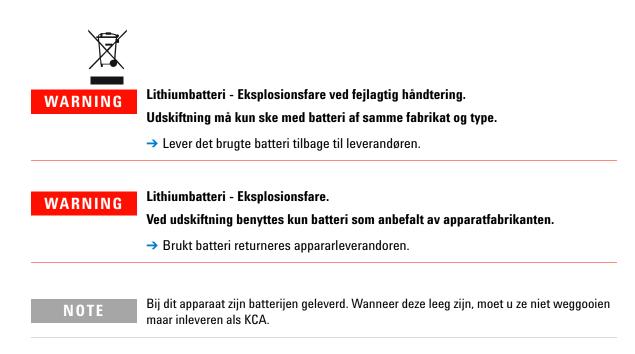
# **Lithium Batteries Information**

# WARNING

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.

Danger of explosion if battery is incorrectly replaced.

- Discharged Lithium batteries shall be disposed off locally according to national waste disposal regulations for batteries.
- → Replace only with the same or equivalent type recommended by the equipment manufacturer.



#### 14 Appendix

The Waste Electrical and Electronic Equipment (WEEE) Directive (2002/96/EC)

# The Waste Electrical and Electronic Equipment (WEEE) Directive (2002/96/EC)

#### Abstract

The Waste Electrical and Electronic Equipment (WEEE) Directive (2002/96/EC), adopted by EU Commission on 13 February 2003, is introducing producer responsibility on all Electric and Electronic appliances from 13 August 2005.

# NOTE



This product complies with the WEEE Directive (2002/96/EC) marking requirements. The affixed label indicates that you must not discard this electrical/electronic product in domestic household waste.

Product Category: With reference to the equipment types in the WEEE Directive Annex I, this product is classed as a "Monitoring and Control instrumentation" product.

Do not dispose off in domestic household waste

To return unwanted products, contact your local Agilent office, or see www.agilent.com for more information.

# **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**

# **Use of Solvents**

Observe the following recommendations on the use of solvents.

- · Brown glass ware can avoid growth of algae.
- 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 sulfuric acid and nitric acid, especially at higher temperatures (if your chromatography method allows, replace 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<sub>3</sub> + O<sub>2</sub>  $\rightarrow$  2COCl<sub>2</sub> + 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,
- Solvents containing strong complexing agents (e.g. EDTA),
- Mixtures of carbon tetrachloride with 2-propanol or THF.

# Use of Solvents with Agilent 1260 Infinity Bio-inert LC systems

Observe the following recommendations on the use of solvents with Agilent 1260 Infinity Bio-inert LC systems.

- Brown glass ware can avoid growth of algae.
- Small particles can permanently block capillaries and valves. Therefore always filter solvents through 0.4  $\mu m$  filters.

Do not use solvents or samples which are not compatible with materials in the flow path (see in this manual "Bio-inert materials" and "Solvent information for parts of the 1260 Infinity Bio-inert LC system".

#### Appendix 14 Agilent Technologies on Internet

# **Agilent Technologies on Internet**

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 modules for download.

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# In This Book

This manual contains technical information about the Agilent 1260 Infinity Bio-inert High Performance Autosampler G5667A.

- introduction and specifications,
- installation,
- using and optimizing,
- troubleshooting and diagnose,
- maintenance,
- parts identification,
- hardware information,
- safety and related information.

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