Agilent InfinityLab LC Series

1290 Infinity III High-Dynamic-Range DAD Solution

User Guide

Notices

Document Information

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Contents

In This Book

This guide covers the Agilent 1290 Infinity III High-Dynamic-Range DAD Solution.

Find detailed information on stack configurations and leak and waste handling in the manuals of the modules, that are necessary to run this solution.

1 What is the HDR-DAD Solution?

This chapter provides an overview of HDR-DAD and its intended use.

30x Wider linear UV-Range – Quantification of Widely Different Concentrations in a Single Run

The typical linear range of around $1 \cdot 10^5$ for UV-detection is not sufficient for the analysis of samples or compound mixtures with widely different concentration levels. Usually, these samples require additional sample preparation steps (dilution/concentration), in combination with a second injection and sometimes recalibration.

The HDR-DAD solution eliminates these sample preparation steps. It increases the linear range by a factor of 30 and allows to acquire all sample information in one single assay. It significantly reduces the turnaround time and increases the productivity.

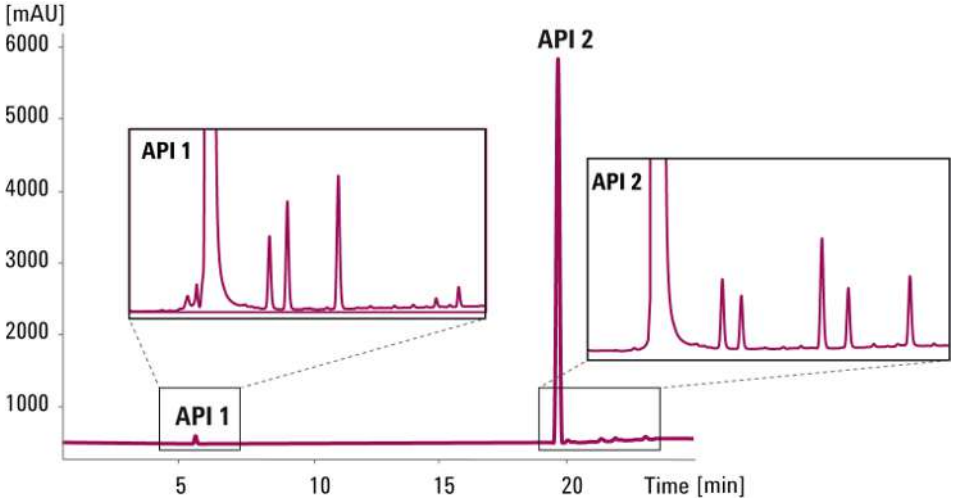


Figure 1: The HDR-DAD solution enables quantification of widely different concentrations in one run

This is achieved by Agilent's Max-Light flow cells. Optofluidic waveguides in these flow cells facilitate total internal reflection for superior light transmission. This significantly reduces the detector noise regardless of the length of the flow path.

The HDR-DAD solution combines two Agilent Diode Array Detectors (possible combinations: G4212A/B x G4212A/B or G7117A/B x G7117A/B); one is fitted with a 3.7 mm path length flow cell, while the other has a longer 60 mm path

length flow cell. The signal from the longer path length cell quantifies the low concentrations and the signal from the shorter path length cell is used for the high concentrations. Special algorithms correct the different retention times and compute these signals to one HDR signal. Compared with an earlier generation Agilent diode array detector, the HDR signal covers a 30 times higher linear dynamic range.

Achieve up to 30-fold greater UV-sensitivity

The Agilent HDR-DAD solution extends the upper linearity level nearly three-fold – from 2 to typically 6 absorbance units – which allows you to inject three-fold more sample and simultaneously benefit from 10 -fold lower detector noise. This solution gives you up to 30 -fold greater sensitivity for simultaneous analysis and quantification of main compounds and impurities. You gain more confidence in automated peak integration and achieve greater area precision for trace-level components.

HDR-DAD Solution Data-Representation

The HDR-DAD solution combines two Agilent Diode Array Detectors (possible combinations: G4212A/B x G4212A/B or G7117A/B x G7117A/B). The two detectors transfer their signals to the software. The algorithm of the HDR-DAD solution combines and presents all signals to the user.

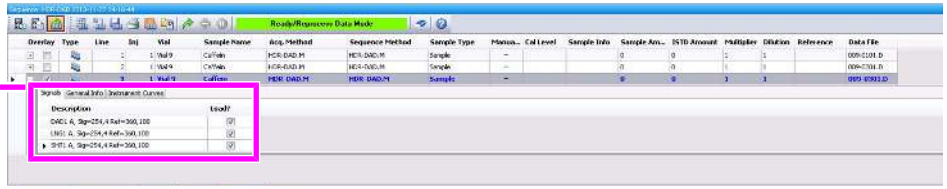


Figure 2: Signal representation in the HDR-DAD solution

It is important to know, which name corresponds to which signal, see Table 1 Data representation in the HDR-DAD solution on page 8 .

Table 1: Data representation in the HDR-DAD solution

Signal name	Signal
DAD1 A	HDR (computed by the weighting function algorithm of the HDR-DAD solution)
LNG1 A	Long flow cell of DAD_1
SHT1 A	Short flow cell of DAD_2

NOTE

The HDR-DAD solution is available in ChemStation and EZChrom. Both software solutions provide the same results.
Nevertheless, the two software solutions show different starting points of the HDR-DAD signal:
ChemStation:
The HDR signal is not starting from zero. The starting time of the HDR signal is calculated with the configuration settings of the delay volume between detectors and the linked pump.
EZChrom:
The signal is displayed starting from zero and the delay is present at the end of the chromatogram.
For details, see Table 3 Differential display of delay time in ChemStation and EZChrom on page 43

2 Specifications

This chapter provides information on performance specifications.

Specifications of the 1290 Infinity III High-Dynamic-Range DAD Solution Kit

Table 2: Performance Specifications of the High-Dynamic-Range DAD Solution Kit (G2199AA)

Type	Specification	Comments
Detection type	1024-element photodiode array	
Light source	Deuterium lamp	equipped with RFID tag that holds lamp typical information.
Number of signals	8	
HDR-DAD Maximum sampling rate	80 Hz	
Short term noise	< ± $0.85 \cdot 10^{-6}$ AU/cm Typically < ± $0.60 \cdot 10^{-6}$ AU/cm	at 230 /4 nm, slit width 4 nm, TC 2 s, ASTM
Drift	< $0.5 \cdot 10^{-3}$ AU/h	at 230 nm
Linearity	> 5.4 AU (5 %) Typically 6.7 AU (5 %)	at 265 nm
Wavelength range	190 – 640 nm	
Wavelength accuracy	± 1 nm	self-calibration with deuterium lines
Slit width	Programmable: 1 , 2 , 4 , 8 nm Fixed slit: 4 nm	(with G7117B, G4212A) (with G7117A, G4212B)
Diode width	~ 0.5 nm	
Wavelength bunching	2 – 400 nm	programmable in steps of 1 nm
Flow cells	Max-Light Cartridge Cell (60 mm, V(s) 4.0 µL) (G4212-60007) HDR Max-Light Cartridge Cell (3.7 mm, V(s) 0.4 µL) (G4212-60032)	70 bar (1015 psi) Maximum Operating Pressure (MOP) [1] 150 bar (2175 psi) Maximum Incidential Pressure (MIP) [2] cartridge type, equipped with RFID tags that holds cell typical information.
Spectral tools	Data analysis software for spectra evaluation, including spectral libraries and peak purity functions	

Type	Specification	Comments
Analog outputs	Recorder/integrator: 100 mV or 1 V, output range 0.001 – 2 AU, one output	
Communications	G4212A/B: LAN, controller-area network (CAN), RS-232C, APG Remote: ready start, stop and shut-down signals G7117A/B: LAN, controller-area network (CAN), USB, ERI/APG Remote: ready start, stop and shut-down signals	
GLP features	RFID for electronics records of flow cell and UV lamp conditions (path length, volume, product number, serial number, test passed, usage) Early maintenance feedback (EMF) for continuous tracking of instrument usage in terms of lamp burn time with user settable limits and feedback messages. Electronic records of maintenance and errors. Verification of wavelength accuracy with deuterium lines.	
Safety and maintenance	Extensive diagnostics, error detection and display through Agilent Instant Pilot and Agilent Lab Advisor software. Leak detection, safe leak handling, leak output signal for shutdown of pumping system. Low voltages in major maintenance areas.	
Others	Electronic temperature control (ETC) for the complete optical unit	

[1] Maximum Operating Pressure (MOP): The maximum pressure at which the system can operate continuously under normal conditions.

[2] Maximum Incidential Pressure (MIP): The maximum pressure which the system can experience during a short time.

3 Installation of the HDR-DAD Solution

This chapter provides information on installation of the HDR-DAD Solution.

Unpacking the Solution Kit

Ensure all parts and materials have been delivered. The delivery checklist for the 1290 Infinity III High-Dynamic-Range DAD Solution Kit (G2199AA) is shown below.

Qty.	p/n	Description
1	5067-4660	Capillary, SST 0.11 mm ID 220 mm long
1	5063-6524	Caffeine in Water, 10 mg/L, 50 mL
1	5022-2159	Restriction capillary, SST 0.12 mm ID, 2 m long
1	G1680-63721	Network LAN Switch
3	8121-0008	LAN Cable, shielded
1	N/A	HDR-DAD USB dongle Contains licence to enable HDR-DAD solution
1	Power Cord	Part number as ordered (country specific)
1	G2199-90000	Printed manual
1	5188-8049	USB A F-USB Mini B M OTG (Module to Flash Drive) For the installation of the HDR-DAD USB dongle into a G7117A/B

Flow connections and network setup

For details on the installation of the entire stack, the detector and the flow cell, see the manual *Agilent 1200 Infinity Series Diode Array Detectors* or *Agilent 1290 Infinity III Diode Array Detector*.

Parts required

Qty.	p/n	Description
2		Agilent 1290 Infinity II Diode Array Detector The HDR-DAD solution supports all combinations of the two mandatory DADs (G7117A/G7117A, G7117A/G7117B or G7117B/G7117B). OR
2		Agilent 1290 Infinity Diode Array Detector The HDR-DAD solution supports all combinations of the two mandatory DADs (G4212A/G4212A, G4212A/G4212B or G4212B/G4212B).
1	G4212-60007	Max-Light Cartridge Cell (60 mm, V(σ) 4.0 µL)
1	G4212-60032	HDR Max-Light Cartridge Cell (3.7 mm, V(σ) 0.9 µL)
1	G2199AA	Agilent 1200 Infinity HDR-DAD Solution Kit

NOTE

The detector with the long path cell needs to be the first one and the detector with the short cell needs to be the second one.
DAD_1: 60 mm Cell - for low concentrations (high sensitivity)
DAD_2: 3.7 mm Cell - for high concentrations (lower sensitivity)

1 Install the DAD_2 on top of DAD_1

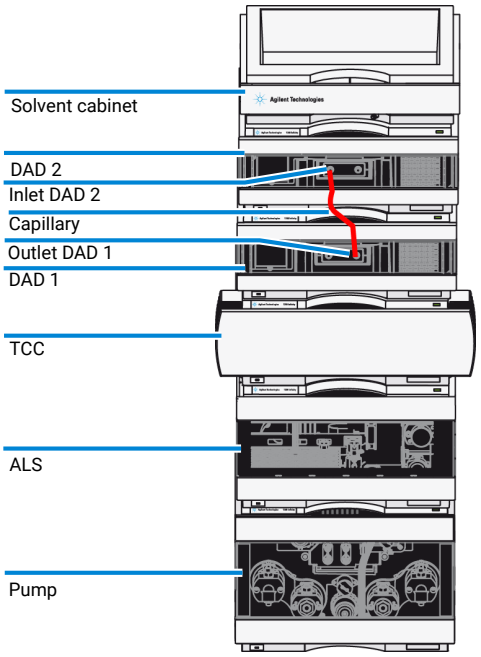


Figure 3: Stack and capillary connection for HDR-DAD (1290 Infinity Stack as an example)

2 Connect the capillaries from the TCC to DAD_1 and the outlet of DAD_1 to inlet the of DAD_2 using Capillary, SST 0.11 mm ID 220 mm long (5067-4660).

3 Setup the network, for an example see Figure 4 on page 16.

NOTE

Connect both detectors to the LAN and in addition connect all the devices via CAN bus.
Configure both LAN connections, otherwise HDR-DAD will remain offline.
Use the included network switch, in case the LC instrument is directly connected to a PC (not via corporate LAN).

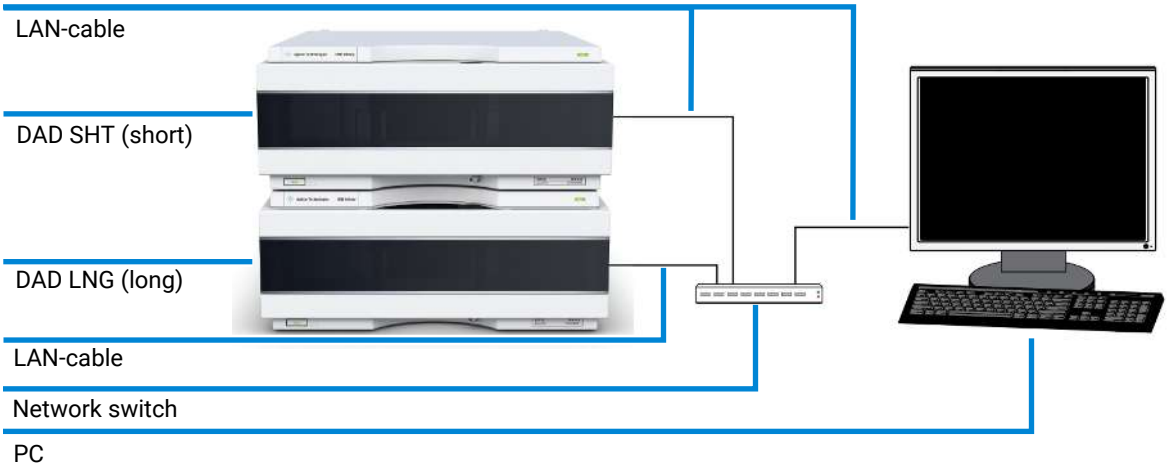


Figure 4: Local LAN connection to a PC (not via corporate LAN)

Enabling the HDR-DAD solution

Parts required

Qty.	p/n	Description
1	N/A	HDR-DAD USB dongle Contains licence to enable HDR-DAD solution
1	5188-8049	USB A F-USB Mini B M OTG (Module to Flash Drive) For the installation of the HDR-DAD USB dongle into a G7117A/B

NOTE

Only one of the DADs needs a license (DAD_1 or DAD_2).

The steps below are valid for the DAD which shall get the license.

1 Turn off the module.

2 Plug-in the HDR-DAD USB dongle into the back of the detector (G7117A/B or G4212A/B).

NOTE You must use the same HDR-DAD USB dongle for reinstallation as was originally used for installation. This, for example, is necessary after exchanging the mainboard of the detector. Thus it's advisable to keep the serial number information of the detector together with the HDR-DAD USB dongle.

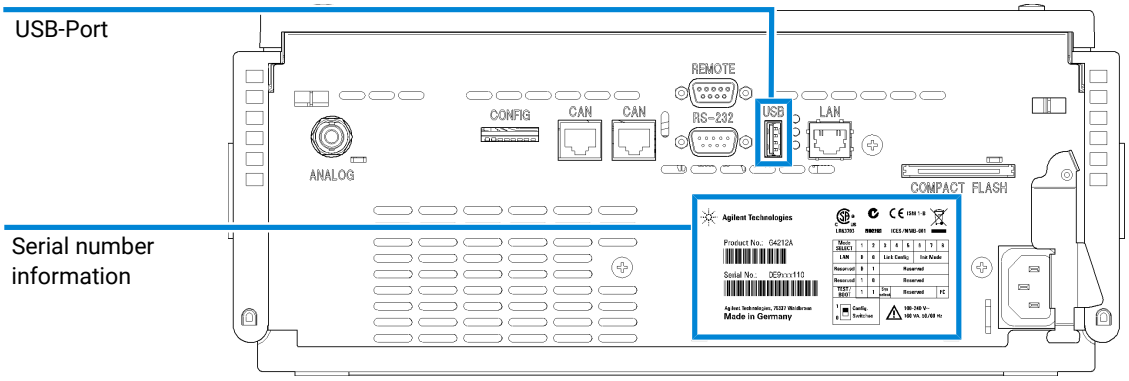


Figure 5: Rear view (G4212A/B)

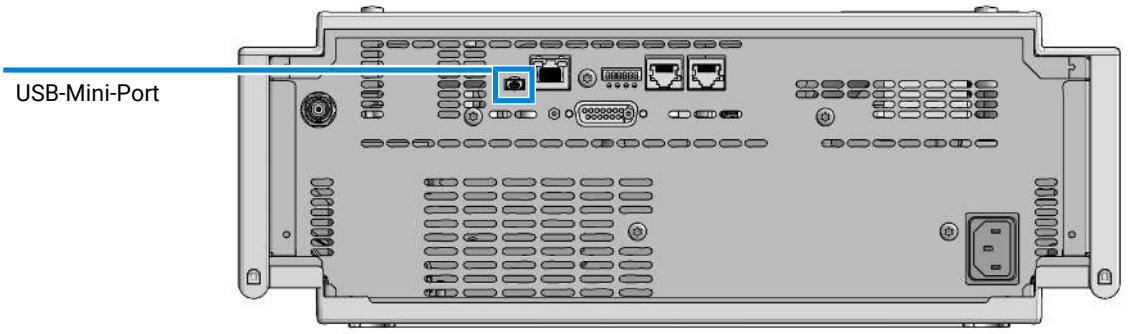


Figure 6: Rear view (G7117A/B)

NOTE For the installation of the HDR-DAD USB dongle into a G7117A/B the 5188-8049 (USB A F-USB Mini B M OTG (Module to Flash Drive)) (part of the HDR-DAD Solution Kit) is required.

3 Turn the module on.

4 Wait until the USB dongle stops blinking.

The HDR-DAD-License is installed on the module.

5 Remove the USB dongle from the module.

The license remains on the module.

Configuring the HDR-DAD Solution

The instrument can be configured via the OpenLAB Control Panel, when setting up the device there.

Software required

- OpenLAB CDS A.01.05 (or higher), ChemStation Edition C.01.05 (or higher), or EZChrom Edition A.04.05 (or higher)
- LC & CE Drivers A.02.09 installed on top
- Firmware (DAD_1 and DAD_2): B.06.57 or higher

Prerequisites

- If the previously available HDR solution was used, uninstall it first.

Auto configuration of the HDR-DAD solution

1 Configure your instrument in the Agilent OpenLAB Control Panel.

2 Click on Configure Instrument.

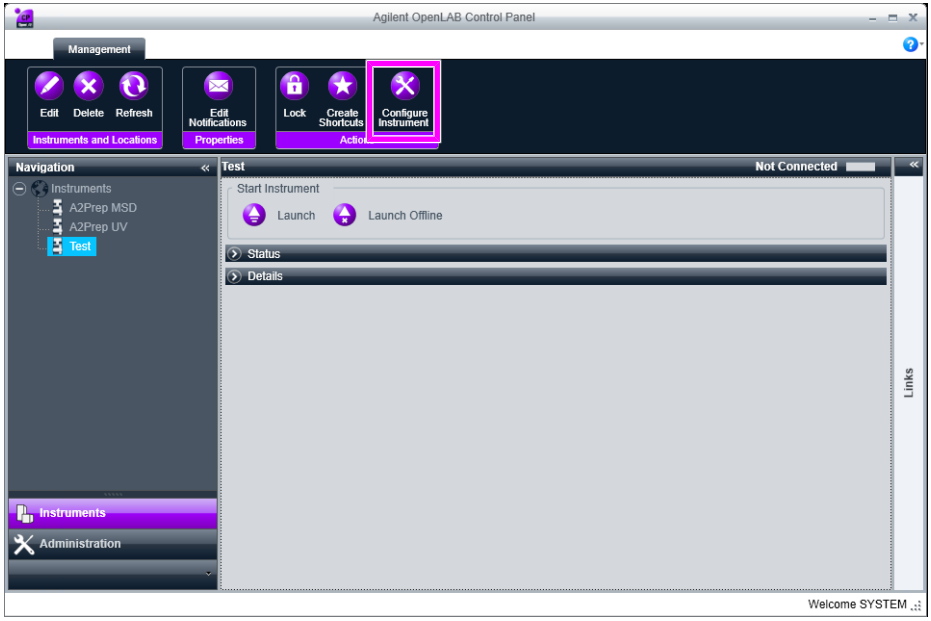
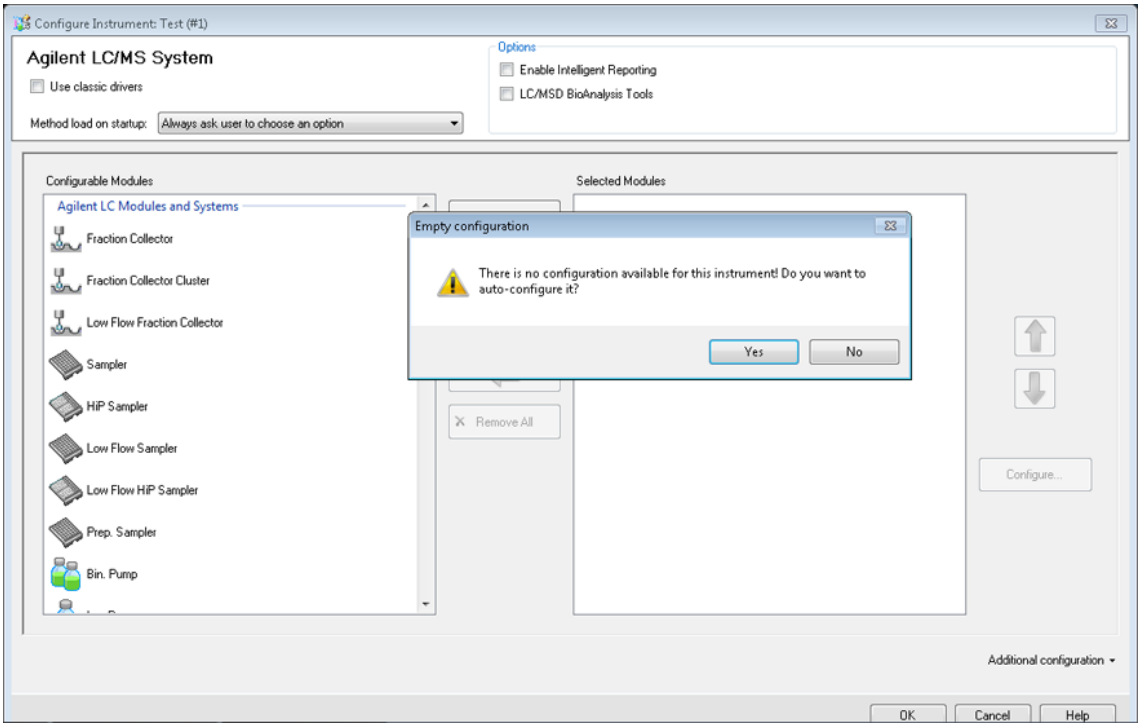
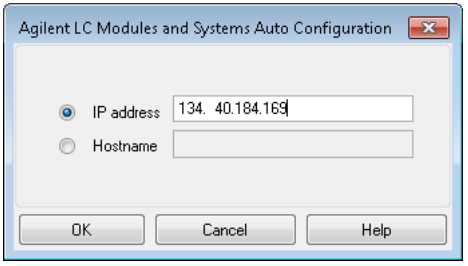


Figure 7: Control Panel of OpenLAB CDS

3 Click **Yes**.



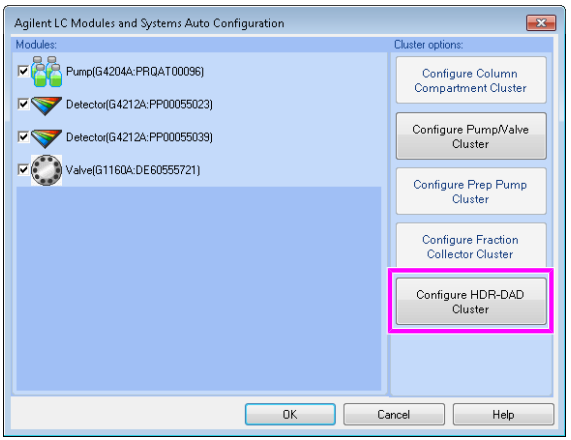
4 Enter the **IP address** or **Hostname** of the DAD with the long cell and click **OK**.



After Auto-Configuration the Post Auto Configuration dialog is shown.

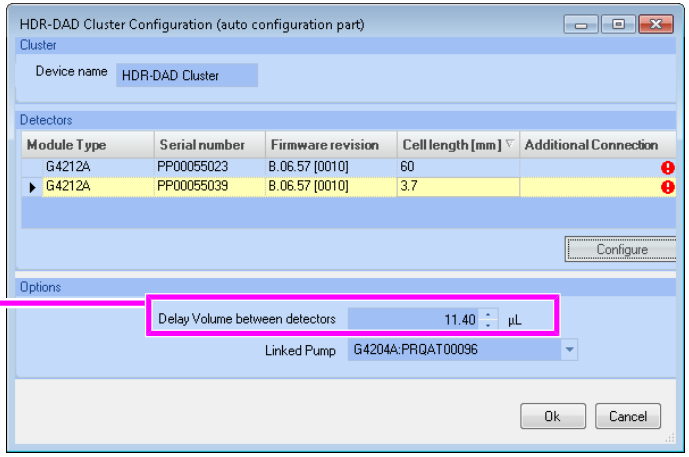
5 To configure the instrument click **Configure HDR-DAD Cluster**.

NOTE In case the prerequisites are not fulfilled, the button is not active. A tooltip informs about the reason.



The detector with the longest cell is always the first detector in the **Detectors** list.

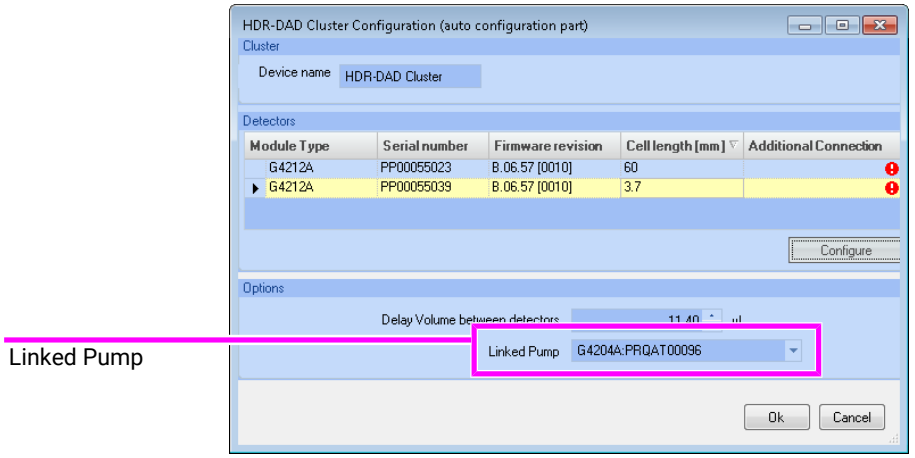
6 Define the **Delay Volume between detectors** (calculate the delay volume upfront, see Calculation of the Delay Volume on page 28).



Delay volume

NOTE The default value as specified in the software is 11.0 µL. Recheck for correct delay compensation in additional runs.

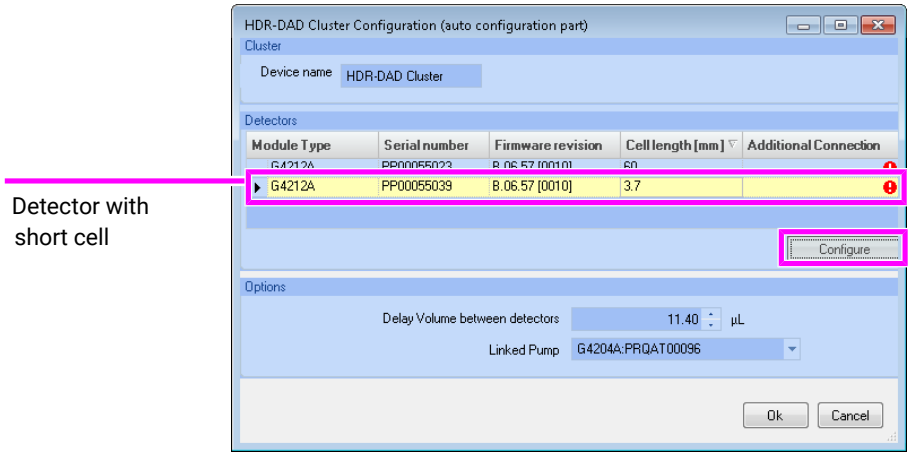
7 Specify the linked pump.



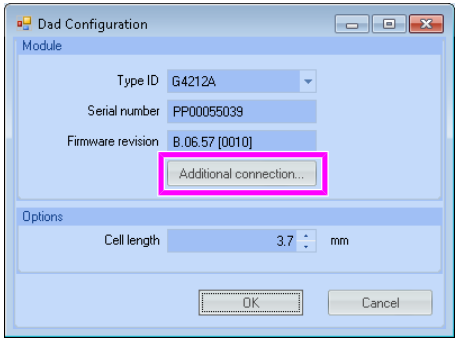
NOTE The **Linked Pump** is used to recalculate the delay for different flow values.

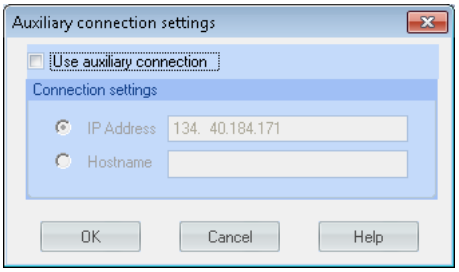
8 Configure the second LAN connection.

a Select the detector with the shorter cell and click **Configure**.

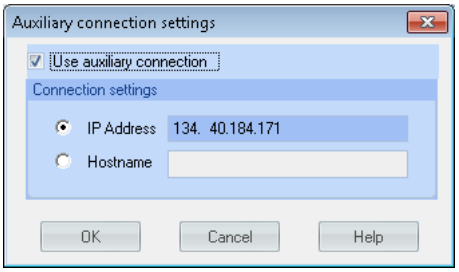


b Click Additional connection...

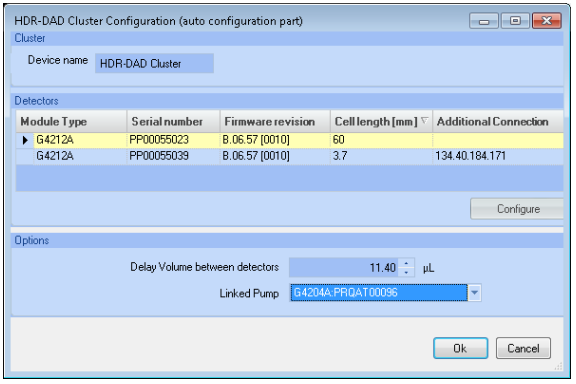




c Check **Use auxiliary connection**, enter **IP Address** or **Hostname** of the detector with the *shorter cell* and click **OK**.



9 In **HDR-DAD Cluster Configuration (auto configuration part)** click **OK**.



The HDR-DAD solution now is configured.

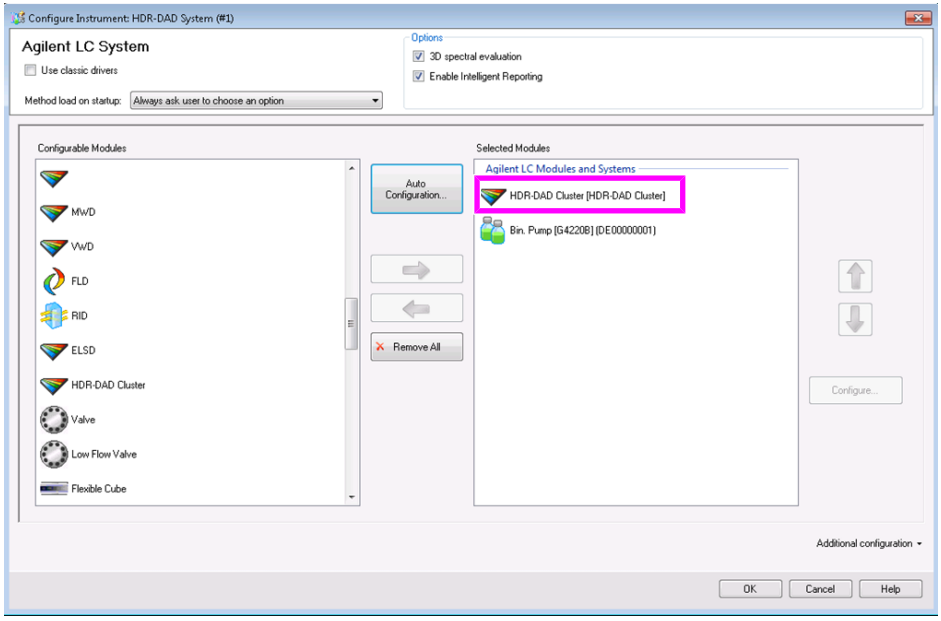


Figure 8: Configured system

NOTE No other detectors are supported together with HDR.

Calculation of the Delay Volume

Parts required

Qty.	p/n	Description
1	5022-2159	Restriction capillary, SST 0.12 mm ID, 2 m long
1	5063-6524	Caffeine in Water, 10 mg/L, 50 mL IQ test sample, for UV/VIS spectrophotometer

No shipment of caffeine standard to China.

NOTE

Suggestion:
For statistical calculations use 6 sample injections.

1 Install 5022-2159 (Restriction capillary, SST 0.12 mm ID, 2 m long) in place of the column.

2 Inject 1 µL of 5063-6524 (Caffeine in Water, 10 mg/L, 50 mL) at a moderate flow rate (for example 0.5 µL/min).

NOTE

The amount of sample is selected to run both detectors in linear range (for example 50 µg/mL, V_{inj}= 1 µL => 50 ... 100 mAU/cm).

3 Calculate the **Delay Volume**.

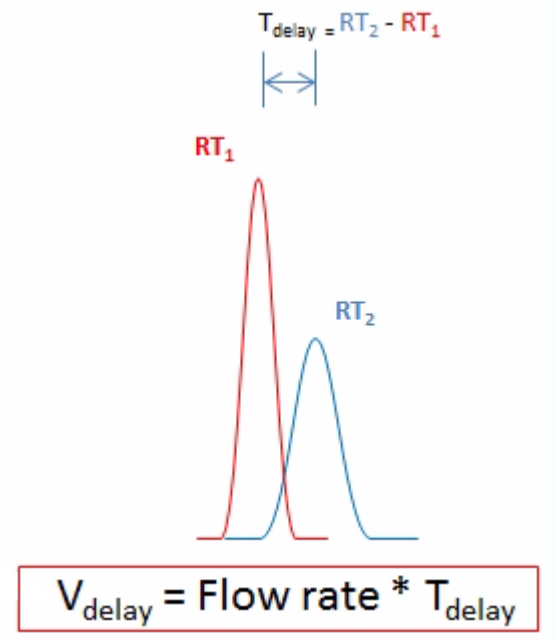
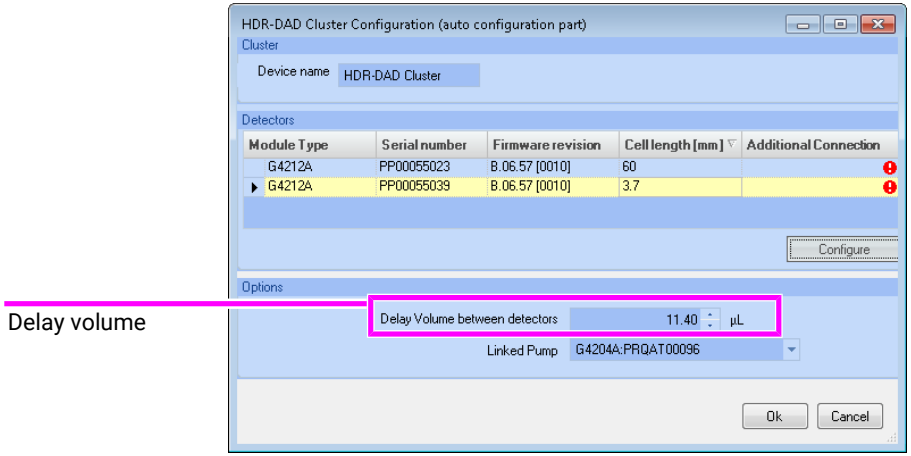


Figure 9: Calculation of the **Delay Volume**

Delay Time: $T_{delay} = RT_{DAD\ 2} - RT_{DAD\ 1}$

Delay Volume: V_{delay} = Flow rate * T_{delay}

4 Open the **High Dynamic Range DAD Configuration**, enter the calculated delay volume and click **OK**.



5 Eventually fine tune the parameter delay volume in further runs.

4 Using the HDR-DAD Solution

This chapter provides information on the main function of the user interface for the HDR-DAD solution. For more details refer to the software manual or the online help.

Status Information and Module Information Dialog of the HDR-DAD Cluster

NOTE

For details on how to use the controller software, see the manual *Agilent 1200 Infinity Series Diode Array Detectors*, *Agilent 1290 Infinity III Diode Array Detector* or the online help.

There are some specialties of the HDR-DAD cluster control panel, compared to the use with a single DAD:

- The DAD_1 (long cell) is always shown on the top line, DAD_2 (short cell) on the 2nd line.

Figure 10: Status of the HDR-DAD Cluster

- The signals shown in the status table of the HDR-DAD Cluster are the calculated HDR signals.

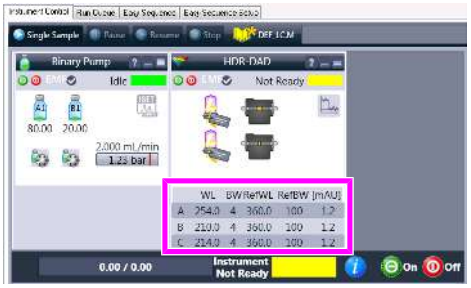


Figure 11: HDR-DAD Signals

- Tooltips show lamp (see Figure 12 on page 33) and cell (see Figure 13 on page 33) tag information for DAD_1 and DAD_2

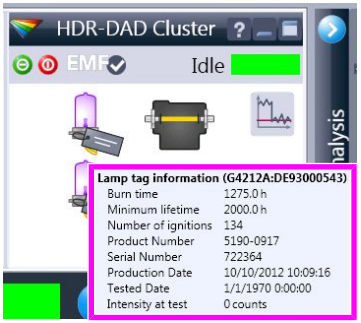


Figure 12: Lamp tag information

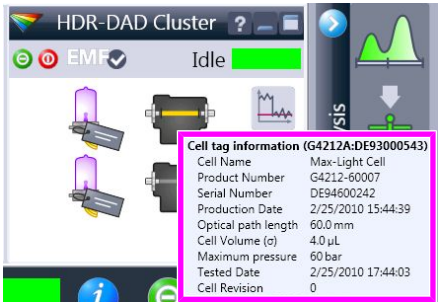


Figure 13: Cell tag information

- The module information dialog shows information about the access point and the additional 2nd LAN connection of the HDR-DAD cluster.

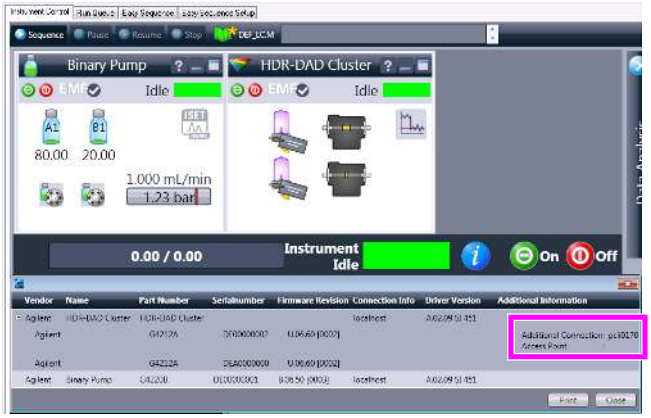


Figure 14: Information on Additional Connection and Access Point

Method Settings in the HDR-DAD Solution

NOTE

The following section describes only settings which are only relevant for the HDR-DAD solution.
For details on method settings as they are important for general use of the DAD, see the manual *Agilent 1200 Infinity Series Diode Array Detectors*, *Agilent 1290 Infinity III Diode Array Detector* or the online help.

The following method setting are relevant for the HDR-DAD solution:

- The **Signals** section provides 8 HDR signals (Signal A-H) with a bandwidth from 190 – 640 nm and its corresponding signals of the single DADs as LNG and SHT (for each detector 8 signals (LNG A-H and SHT A-H)

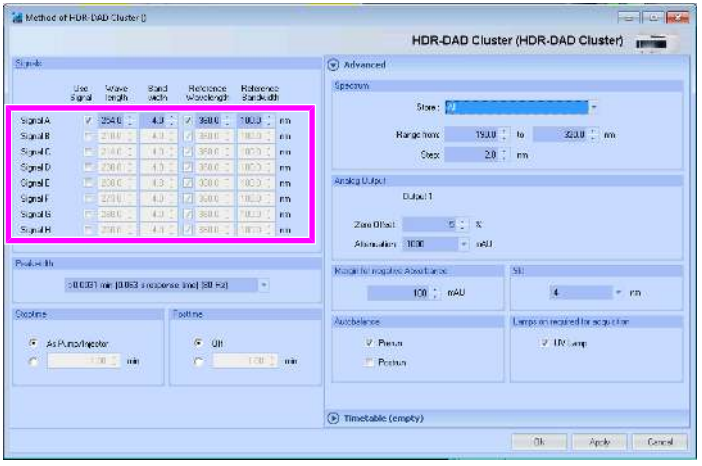


Figure 15: HDR-DAD Bandwidth

- In the **AdvancedSpectrum** section, the user may chose the storage of **All** spectra or **None** from a dropdown list. The HDR-DAD solution supports full HDR spectra data (190 – 640 nm)

  NOTE: There is no support for other spectra modes.Only the **Analog Output** channel of DAD_2 with the short cell delivers the HDR-DAD signal.(The **Analog Output** channel of DAD_1 with the long cell will show 0 V signal.)

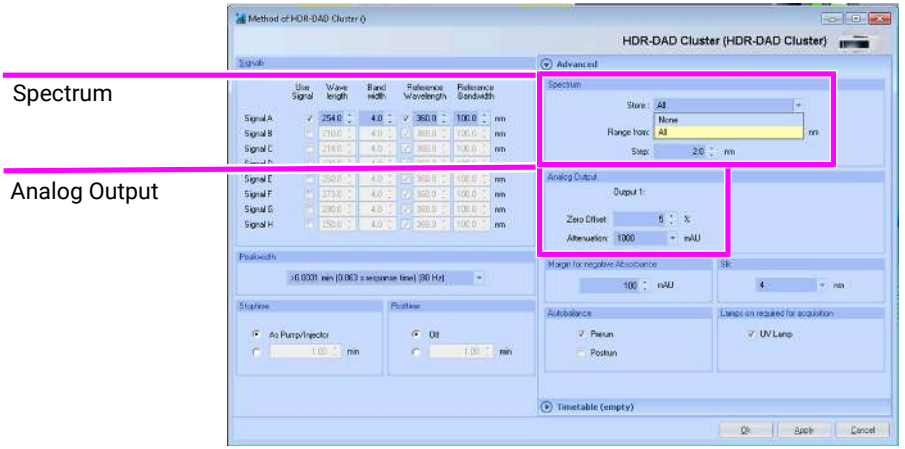


Figure 16: HDR-DAD Spectra support

- Peakwidth

  The data rate is settable from 0.31 – 80 Hz

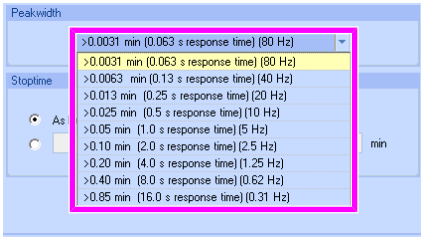


Figure 17: HDR-DAD Peakwidth settings

- Slit

  The slit width is settable to 1 , 2 , 4 , or 8 nm

  NOTE: If one or both detectors are G7117A or G4212B (module has a fixed slit), the slit automatically is set to 4 nm and cannot be adjusted.

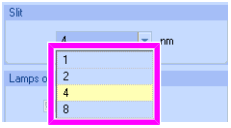


Figure 18: HDR-DAD Slit width settings

Status Display During a run with the HDR-DAD

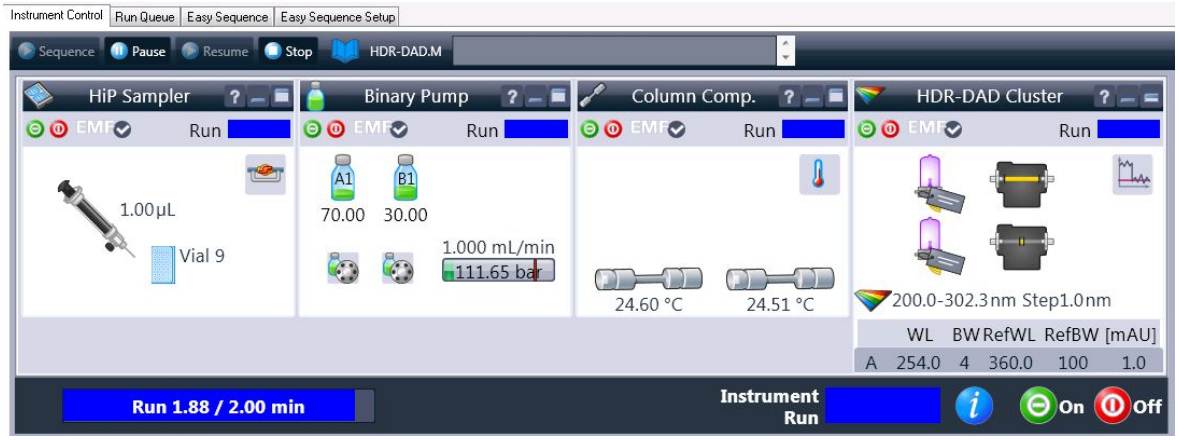


Figure 19: Display during a HDR-DAD run

NOTE

For details on the general functions of the status display, see the manual *Agilent 1200 Infinity Series Diode Array Detectors*, *Agilent 1290 Infinity III Diode Array Detector* or the online help.

Click on the menu items in the context menu of the status dashboard offers the following HDR-DAD solution specific functions:

- Identify Device lets both detectors blink
- Balance triggers balance on both detectors
- Switch On/Switch Off switches lamps of both detectors on/off

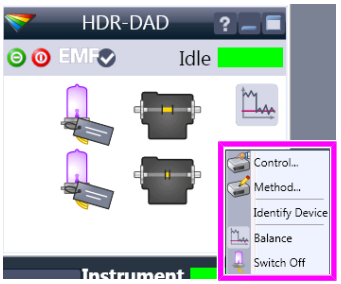


Figure 20: Context menu of the status dashboard

Control User Interface of the HDR-DAD Solution

NOTE

For details on the general functions of the control user interface, see the manual *Agilent 1200 Infinity Series Diode Array Detectors*, *Agilent 1290 Infinity III Diode Array Detector* or the online help.

The following HDR-DAD specific functions exist:

- Selection of radio button **On**/**Off** in section **Lamps**-**UV** turns on/off lamps of DAD_1 and DAD_2

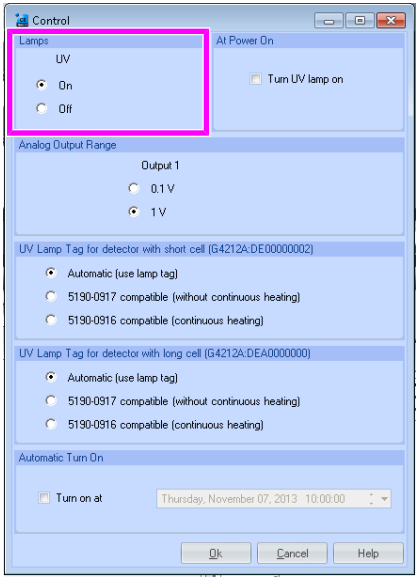


Figure 21: HDR-DAD **Control** User Interface

Method and Run Control User Interface in ChemStation

The **Method and Run Control** user interface provides information on the status of the system.

Figure 22: HDR-DAD solution control screen in ChemStation

NOTE

With EZChrom, the functionality of the HDR-DAD solution is equal and the status dashboard looks akin to ChemStation - but the online plots and spectra look different.

Code for the signals observed:

- The HDR-DAD signals are available as **DAD1A - H** signal
- The original signals of DAD_1 are available as **LNG1A - H** signal (= long cell)
- The original signals of DAD_2 are available as **SHT1A - H** signal (= short cell)

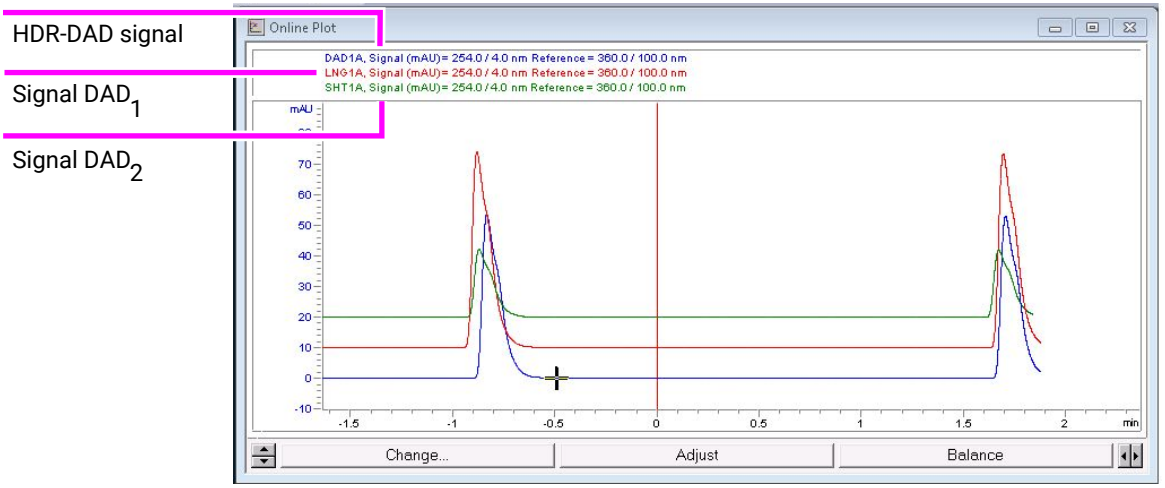


Figure 23: HDR-DAD **Online Plot** section

The HDR-DAD solution also offers the possibility to review the spectra online while acquiring data. The **HDR-DAD Cluster: Spectrum (mAU)** section shows the HDR spectra.

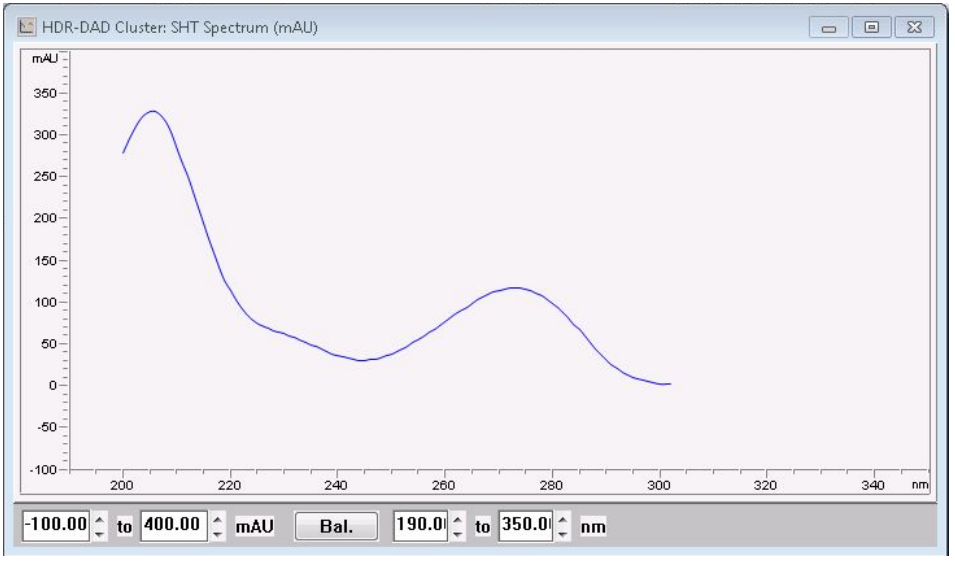


Figure 24: HDR-DAD spectrum

HDR-DAD Solution - Data Analysis

The HDR-DAD solution offers the following sections for **Data Analysis**:

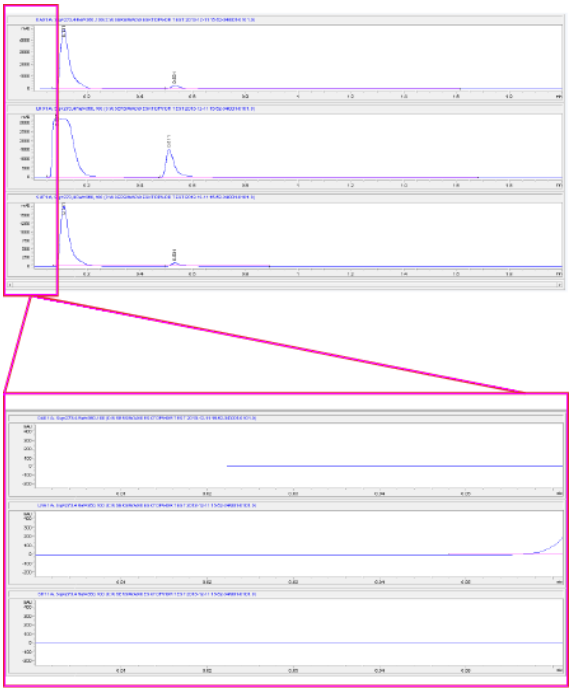
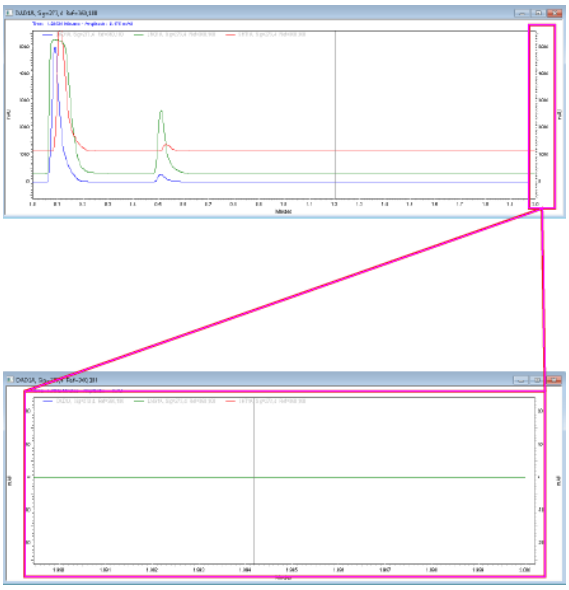
- Three separate signal groups for HDR, DAD_1 (LNG = long cell), DAD_2 (SHT = short cell)

  The user can observe 24 signals 3 (HDR, DAD_1, DAD_2) x 8 signals (A-H)
- Spectra information

NOTE

Display of **Delay Time** in the data analysis software
The flow from DAD_1- capillary - DAD_2 causes a **Delay Time**. The HDR-DAD solution calculates the chromatogram from DAD_1 and DAD_2 signals. Therefore a HDR-chromatogram always starts at earliest with the signal of DAD_2.
In spite of this, ChemStation and EZChrom display the delay time differential.
ChemStation:
The HDR-DAD chromatogram shows a short gap at the beginning of the signal. This reflects the fact that the combined HDR signal can only start as it is delayed by the delay time between the detectors. Whereas the signals of the single detectors start immediately at t = 0 min.
See the ChemStation graphic in Table 3 Differential display of delay time in ChemStation and EZChrom on page 43.
EZChrom:
The HDR-DAD chromatograms always start at t = 0 min and shows a short gap at the end of the signal.
See the EZChrom graphic in Table 3 Differential display of delay time in ChemStation and EZChrom on page 43.

Table 3: Differential display of delay time in ChemStation and EZChrom

ChemStation	EZChrom
	
ChemStation displays a short gap at the beginning of the HDR signal (red rectangle	EZChrom displays a short gap at the end of the HDR signal (red rectangle)

5 Frequently Asked Questions (FAQs)

This chapter answers frequently asked questions.

Frequently Asked Questions

1. What can I do if I observe a leap in the HDR signal?

   Wrong delay calibrations cause leaps in the data signal (see Figure 25 on page 45).

   Double check the delay calibration between the 2 detectors.

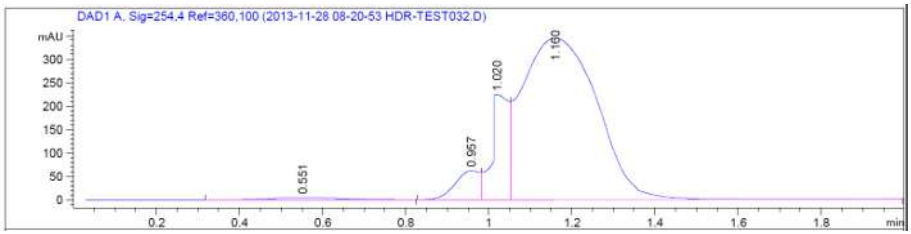


Figure 25: Leaps in the HDR signal

2. How are the signals named?
   - DAD1 = combined HDR-DAD signal
   - LNG1 = signal of the single DAD with the long Max-Light Cartridge Cell installed
   - SHT1 = signal of the other single DAD with the short Max-Light Cartridge Cell installed
3. Can I use additional detectors if I include a UIB II to the HDR-DAD solution?

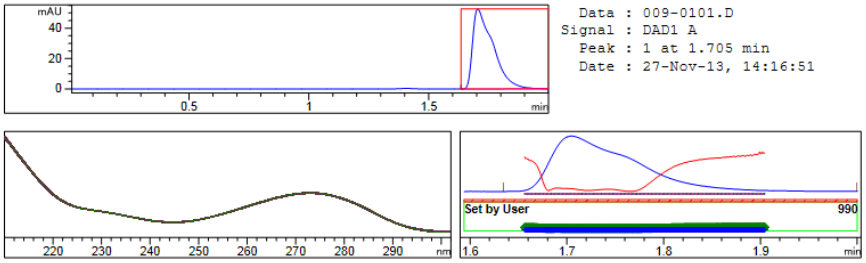
   It is not recommended to use additional detectors with the HDR-DAD solution.
4. Which firmware is requiered to run the HDR-DAD solution?

   You need firmware revision B.06.57 or later to run the HDR-DAD solution.
5. How can I print spectra information in OpenLAB CDS ChemStation edition?

   To print the correct spectra information, adhere to the following procedure:
   - Open Signal Details... dialog (in ChemStation via Calibration > Signal Details...).
   - Make sure that only the DAD 1A signal is loaded (and not any of the LNG1 or SHT1).
   - Open the View > Preferences... dialog, select Signal > Review Options and check Load using signal details

- Save the method and reload the data.
- Now the **Full Report** will contain the correct information whenever printed.

```
Purity results peak 1 at 1.705 min. name : ?
```




```
-> The purity factor is within the threshold limit. <-

Purity factor : 999.798 (1193 of 1193 spectra are within the threshold limit.)
Threshold     : 990.000  (Set by user)
Reference     : Nearest baseline spectrum (stored)  (0.011)
Spectra       : 1193  (Selection automatic, All)

Peak spectra
 1  DAD1, 1.656 (5.0 Fl,Bln) Ref= 0.011
 2  DAD1, 1.657 (5.3 Fl,Bln) Ref= 0.011
```

In This Book

This user guide contains technical reference information about the Agilent 1290 Infinity III High-Dynamic-Range DAD Solution.

The manual describes the following:

- Introduction
- Specifications
- Installation
- Using the HDR-DAD Solution
- Frequently Asked Questions

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