

Agilent 1260 Infinity Bio-inert Quaternary LC

Features, Technical Details, Applications and **Specifications**

Agilent 1260 Infinity Bio-inert Quaternary LC

The Agilent 1260 Infinity Bio-inert Quaternary LC system is a dedicated solution for large bio-molecule analysis. The design of new metal-free components in the sample flow-path and the absence of iron and steel in solvent delivery ensures the integrity bio-molecule, minimizes unwanted surface interactions and increases column life-time. This is ideal when working under harsh solvent or pH conditions. The power ranges from lowest pressure for traditional bio-purification columns up to high pressure STM analytical bio-columns. Together with the new Bio-HPLC column portfolio for SEC and IEX and 10x higher sensitivity, highest resolution per time is achieved for protein and NBE characterization.

Features

- · Complete metal-free sample contacting surfaces
- · Mobile phase wetted parts iron and steel free in solvent delivery unit
- Low and high-pressure column capability (0 600 bar)
- 10x higher sensitivity
- High salt tolerance (2M) and wide pH range (1-13, short term 14)
- · Active seal wash included
- Quaternary solvent blending
- Analytical and semi-prep up to 10 mL/min with inert fraction collection
- · Inert flow-cells for UV and fluorescence detection
- · Inert solvent and column selection valves
- · Novel capillary and connection design
- · Characterization of bio-therapeutics
- · Large portfolio of Bio-HPLC columns for SEC, IEX, reversed phase and peptide mapping



Agilent Technologies

Technical Details – Agilent 1260 Infinity Bio-inert Quaternary LC

Routine bioanalysis and biopurification at RRLC performance

The Agilent 1260 Infinity Bio-inert Quaternay LC System can withstand harsh conditions for bio-analytic and biopurification applications and maintain the performance of a modern UHPLC instrument. This is possible through the careful design of all components that are in contact with mobile phase and sample. Problems often associated with large bio-molecules are unspecific surfaces interaction , analyte discrimination and metal ion release which can cause decreased column lifetime, peak-tailing or lack of resolution. In addition high salt concentration and extreme pH values cause corrosion and decrease robustness and instrument uptime. These problems required special care and tedious passivation procedures which are no longer necessary with the Agilent 1260 Infinity Bio-inert Quaternary LC system thus increasing throughput and efficiency of your bioanalytical or semi-prep biopurification lab.

Routine bioanalysis and biopurification at RRLC performance

- · Ceramic injector needle and PEEK sample loop protected by high pressure housing (autosampler)
- · 600 bar pressure-stable PEEK needle seat and capillary connection design (autosampler)
- Metal cladded PEEK (inside) capillaries to warrant high pressure capability and inertness over complete flow path (system)
- · Newest socket design for unions and fittings to ensure biocompatibility as well as high pressure capability (system)
- Metal-cladded PEEK capillaries for the clip in heating elements in the column oven (TCC)High pH stability and biocompatible detector flow cell containing PEEK capillaries, fittings and ferrules, sapphire window and ceramic detection cell body (MWD or DAD)
- Inert PEEK capillaries for high sensitivity DAD flow cell and Fluorescence detector cell (detector)
- · Biocompatible analytical scale fraction collection (fraction collector)
- Titanium capillaries, damping and mixing system in solvent delivery module (pump)

Applications – Agilent 1260 Infinity Bio-inert Quaternary LC

Characterization of bio-therapeutics

Physico-chemical characterization and confirmation plays a crucial role in the New Biological Entity (NBE) and biotherapeutics workflow to ensure drug safety and efficacy. Agilent offers a broad spectrum of tools for all assays in order to fulfill the regulatory requirements. For the LC-based tests the Agilent 1260 Infinity Bio-inert Quaternary solution offers a high degree of flexibility to address the needs of a quality control environment as well as method development flexibility for your applications in SEC, ion-exchange, peptide mapping, confirmation and glycan analysis.Therapeutic monoclonal antibodies (mABs) are characterized by a variety of assays to warrant drug safety and efficacy. The Agilent 1260 Infinity Bio-inert Quaternary solution is a flexible tool addressing the major requirements.

Application Examples

Ion-exchange to detect truncation, deamidation and oxidation and glycosylation often represented by charge state variant

With the Agilent 1260 Infinity Bio-inert Quaternary LC and the polymeric BioMab WCX columns Agilent offers a unique single vendor solution for highest performance requirements. The combination of column technology from 10 µm particles to 1.7 µm particles and biocompatibility results in increased robustness, superior resolution and prolonged column lifetime.



Analysis of charge variants of a mAB.

Size exclusion chromatography for purity and aggregation analysis

With the Agilent 1260 Infinity Bio-inert Quaternary LC and Bio SEC 5 and Bio SEC 3 columns, robust performance and high reproducibility under different buffer conditions with or without detergents is obtained. In combination with a variety of detectors such as UV and fluorescence impurities can be easily resolved and detected with superior sensitivity.



Peptide Mapping

The Agilent 1260 Infinity Bio-inert Quaternary LC offers RRLC capability combined with low surface activity especially for critical samples. In combination with the Agilent Eclipse Plus 1.8 µm particle columns or with the Poroshell 120 stationary phases, high resolution and superior peak capacities are achieved in order to confirm the identity of the analyzed NBE drug in a QA/QC environment.



Peptide map of a therapeutic protein on Eclipse Plus C18 3.0 x 100 mm, 1.8 μm, flow rate 0.8 mL/min.

Specifications – Agilent 1260 Infinity Bio-inert Quaternary Pump (G5611A)

Туре	Specifications
Hydraulic system	Dual plunger in series pump with proprietary servo-controlled variable stroke drive, floating plungers and active inlet valve, integrated 4-channel degassing unit
Setable flow range	0.001 – 10 mL/min, in 0.001 mL/min increments
Recommended flow range	0.2 – 10.0 mL/min
Flow precision	< 0.07% RSD, or < 0.02 min SD whatever is greater, based on retention time at constant room temperature
Flow accuracy	$\pm1\%$ or 10 $\mu L/min$ whatever is greater
Pressure	Operating range 0 $-$ 60 MPa (0 $-$ 600 bar, 0 $-$ 8700 psi) up to 5 mL/min Operating range 0 $-$ 20 MPa (0 $-$ 200 bar, 0 $-$ 2950 psi) up to 10 mL/min
Pressure pulsation	<2 % amplitude (typically <1.3 %), at 1 mL/min isopropanol, at all pressures >1 MPa (10 bar)
Compressibility compensation	User-selectable, based on mobile phase compressibility
Recommended pH range	1.0 – 13, short term 14
Gradient formation	Low pressure quaternary mixing/gradient capability using proprietary high-speed proportioning valve Delay volume 600 – 800 µL, dependent on back pressure
Composition range	0 – 95% or 5 – 100%, user selectable
Composition precision	< 0.2% RSD, at 0.2 and 1 mL/min
Control and data evaluation	Agilent control software (e.g. ChemStation, EZChrom, OL.)
Communications	Controller-area network (CAN), GPIB, RS-232C, APG Remote: ready, start, stop and shut-down signals, LAN optional
Wetted materials	Titanium, Gold, Platin-Iridium, Sapphire, PEEK, PEKK, PTFE
Active seal wash	Included

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

Туре	Specifications
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	Leak detection and safe leak handling, low voltages in maintenance areas, error detection and display
Injection range	$0.1-100~\mu$ L in 0.1 μ L increments. Up to 40 μ L with reduced injection volume kit (hardware modification required). Up to 1500 μ L with multiple draw (hardware modification required)
Precision	Typically < 0.25% RSD from 5 – 100 μ L. Typically < 0.5% RSD from 2 – 5 μ L. Typically < 0.7% RSD from 1 – 2 μ L volume. Measured with injections of benzylalcohol
Pressure range	Up to 600 bar (8700 psi)
Sample viscosity range	0.2 – 5 cp
Sample capacity	2 x well plates (MTP) + 10 x 2 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 (0.5/1.5/2.0mL) in 2 x 27 Eppendorf tube plates
Injection cycle time	Typically < 17 s using the following standard conditions: Default draw speed: 100 μ L/min. Default eject speed: 100 μ L/min. Injection volume: 5 μ L
Carry-over	Typically < 0.004% using the following conditions:
	Column: Agilent ZORBAX SB-C18, 21 x 50 mm 1.8 μm
	Mobile Phase: A: H ₂ 0 + 0.05 % TFA, B: ACN+ 0.045 TFA
	Gradient: 0.1 min 10% B, 3.1 min 90% B, 3.2 min 90 % B, 3.21 10 %B, 4.5 min stop
	Flow rate: 0.5 mL/min
	Temperature: 25 °C
	Wavelength: 257 nm
	Sample: 1200 ng/ μ L Chlorhexidine (disolved in H ₂ O with 0.1% TFA), 1 μ L injected and measured both on Agilent 6410 QQQ and Agilent DAD (G1315C). Wash solution: H ₂ O with 0.1% TFA (5 sec)
pH-range	1-13 (short term 14)

Materials used in sample flow path	
Needle seat	PEEK
Needle	Ceramic
Sample loop	Metal-cladded (outside) PEEK (inside)
Injection valve	PEEK, ceramic, FFKM
Other	PPS02, PTFE
Sample cooling	Optional with G1330B 4 $^{\circ}$ C – 40 $^{\circ}$ C

Specifications – Bio-inert LC capillaries, connectors, detector flow cells, solvent heating elements and valves

Capillaries and connectors (complete system)		
Materials (flow path)	Metal cladded (outside), PEEK, PTFE inside surfaces	
pH range	1-13 (short term 14)	
Maximum pressure	600 bar	
DAD /MWD flow cells G5615-60022 for G1315 C/D and G1365 C		
Materials (flow path)	PEEK, saphire window	
pH range	1-13 (short term 14)	
Diode array detector G4212A with bio-inert flow cells 10 mm G4212-60008 or 60 mm G4212-60007 high sensitivity flow cells		
Materials (flow path)	PEEK, fused silica	
pH range	1-12	
Fluorescence detector G1321B with bio-inert flow cell G5621-60005		
Materials (flow path)	PEEK, fused silica	
pH range	1-12	
Bio-inert heating elements G5616-60050 (9 μL) for G1316C and G1316A		
Materials (flow path)	PEEK	
pH range	1-13 (short term 14)	
Stand-alone bio-inert valves in G1170A		
Universal Actuator	12/13 solvent selection valve head Bio-inert for up to 12 solvents $$ G4235A $$	PEEK, 210 bar max.
	2-position/6-port G5631A	PEEK/ceramic 600 bar max.
	4 column selection G5639A	PEEK/ceramic 600 bar max.

Specifications – Agilent 1260 Infinity Analytical Bio-inert Fraction Collector (G5664A)

Туре	Specifications
Delay volume	Approximately 50 µL
Maximum system flow	10 mL/min
Fraction containers	Shallow or deep well plates up to 48 mm height (96 or 384 format)
	Test tubes up to 48 mm height
	Autosampler vials (2 mL and 6 mL)
	Eppendorf safe-lock tubes (0.5 mL, 1.5 mL , 2.5 mL)
Fraction trays	Full trays (cover complete fraction collector)
	4 well plates
	40 x 20 mL test tubes (30 mm od, 48 mm height)
	60 x 15 mL test tubes (25 mm od, 48 mm height)
	126 x 8 mL test tubes (16 mm od, 48 mm height)
	215 x 5 mL test tubes (12 mm od, 48 mm height)
	2 well plates + 10 x 2 mL vials (+ one possible additional half tray)
	100 x 2 mL vials (+ one possible additional half tray)
	Half trays (up to 3 per fraction collector)
	15 x 6 mL vial
	40 x 2 ml vial

Specifications – Agilent 1260 Infinity Analytical Bio-inert Fraction Collector (G5664A) *(continued)*

Туре	Specifications
Fraction trays	Tube plates for well plate trays (2 or 4 per fraction collector depending on well plate tray)
	Eppendorf safe-lock tubes (27 x 0.5 mL, 1.5 mL or 2.5 mL)
	24 test tubes (18 mm OD)
	54 x 2 mL vial
	15 x 6 mL vial
Cooling	Optional
Trigger modes	Time slices and peak (threshold, up-/downslope, upper threshold and timetable), Boolean logic for different detector signals, combination of different modes, manual trigger (supported by 1200 Series Instant Pilot)
Trigger source	Agilent 1200 Infinity Series VWD, MWD and DAD detectors, Agilent 6100 Series Quadrupole LC/MS, ELSD, FLD, RID, third party detectors (require UIB)
Diverter valve	3/2 valve, switching time < 100 ms
Max. pressure	6 bar
Environment	4 – 55 °C constant temperature, < 95% humidity (non-condensing)
Wetted parts	PEEK, ceramic, PTFE

Specifications – Agilent 1260 Infinity Bio-inert Manual Injector (G5628A)

Туре	Specifications
Injection valve	7 port, long-life loops
Loops	20 µL (standard)
	5 μL-5mL (optional)
Wetted parts	PEEK, PTFE, ceramic

www.agilent.com/chem/bio-inert

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