

Keep Your CQA Analysis Biocompatible from Start to Finish

PEEK-lined columns and bio-inert supplies to complement
the Agilent 1290 Infinity II Bio LC system





Confidently Measure the Efficacy and Stability of Your Therapeutic Proteins with Agilent Consumables and InfinityLab Instruments

Biotherapeutic proteins are highly complex molecules, which typically are produced by fermentation using recombinant methodologies. This production process, however, results in the generation of many different variants of these proteins. Ensuring the quality of such materials is paramount. This means confirming the product is correctly manufactured, any impurities are identified and quantified, and the potency of the protein is determined.

As a result, it is necessary to perform tests on the intact, nondenatured molecule. Something as large as a monoclonal antibody may contain more than 1,300 individual amino acids and have a mass of more than 145,000 Daltons. However, identifying a single minor impurity, such as deamidation of asparagine resulting in a mass difference of just one Dalton, is challenging. Only by breaking down the molecule into fragments, and then into smaller polypeptide chains through enzymatic treatment, is it possible to begin to pinpoint some of these subtle differences.

Many types of variants can be created and these are often referred to as post-translational modifications (PTMs), which arise after the protein has been expressed, and can be a consequence of the manufacturing conditions or exposure to conditions that cause changes to occur. Fluctuations in temperature, pH, concentration, or exposure to enzymes can all lead to the development of variants.

The Agilent 1290 Infinity II Bio LC system—together with PEEK-lined, bio-inert Agilent AdvanceBio columns and standards—can help you characterize PTMs and other critical quality attributes (CQAs). For molecules that are susceptible to metal-induced artifacts, we have also designed a biocompatible flow path. Importantly, this includes PEEK-lined columns, because columns are the single largest contributor of metal to the flow path.

Be Agilent Sure in Your CQA Monitoring

Agilent AdvanceBio LC columns are designed and produced to provide results you can be sure of when analyzing highly complex biotherapeutic molecules and monitoring their purity, potency, and other critical quality attributes.

To learn more, click the blue, green, and gray boxes.

Titer Determination	Aggregate Analysis	Intact Purity & PTM Analysis		Peptide Mapping & PTM Analysis	Charge Variant Analysis	Glycan Analysis	Amino Acid / Cell Culture Media Analysis	
Affinity	Size Exclusion	Reversed Phase > 150 Å	Hydrophobic Interaction	Reversed Phase < 150 Å	Ion Exchange	Hydrophilic Interaction	Reversed Phase < 150 Å	Hydrophilic Interaction
	AdvanceBio SEC 1.9 µm PEEK	PLRP-S 1000 Å 5 µm PEEK		AdvanceBio EC-C18 PEEK	Bio mAb / Bio IEX NP5 PEEK			AdvanceBio MS Spent Media PEEK
Bio-Monolith Protein A	AdvanceBio SEC 1.9 µm	PLRP-S	AdvanceBio HIC	AdvanceBio Peptide Mapping	Bio mAb	AdvanceBio Glycan Mapping	AdvanceBio Amino Acid Analysis (AAA)	
Bio-Monolith Protein G	AdvanceBio SEC 2.7 µm	AdvanceBio RP mAb 450 Å		AdvanceBio Peptide Plus	Bio IEX (SAX, WAX, SCX, WCX)		ZORBAX AAA	
	Bio SEC-3	ZORBAX RRHD 300 Å, 1.8 µm		ZORBAX 300 Extend-C18	PL SCX, SAX			
	Bio SEC-5	ZORBAX 300SB 3.5, 5, & 7 µm			Bio-Monolith (QA, DEAE, SO3)			
	ProSEC 300S	Poroshell 300 5 µm						
	ZORBAX GF250 & GF450							

Key



Stainless steel (SS) column hardware



Solid PEEK or PEEK-lined bio-inert column hardware



Learn more at www.agilent.com/chem/advancebio

Meet Your Most Complex Biopharma Challenges

Agilent offers robust, reliable solutions for analyzing purity, potency, and other critical quality attributes (CQAs). For molecules that are susceptible to metal-induced artifacts, we also have designed a biocompatible flow path from start to finish.



Biocompatible valve automation

InfinityLab Quick Change valves enable sample enrichment and cleanup, automated column regeneration, and enhanced system flushing.

Built-in sampling versatility

The 1290 Infinity II Bio multisampler allows for adaptable injection volumes, thermostating of temperature-sensitive bio-analytes, and ultralow carryover.



Multiple detection options

Choose from several optical detection capabilities with varying flow cells, including variable wavelength, diode array and fluorescence detectors, and the bio-MDS system.



Versatile column handling

The 1290 Infinity II MCT, featuring the Thermal Equilibration Device, gives high-temperature stability for the most challenging applications.

Various bio-inert heat exchangers, Quick Change and Quick Connect capillaries, fittings, and kits are also available.

Reliable solvent delivery

Built for robustness under high-salt or high-/low-pH conditions, the 1290 Infinity II Bio high-speed pump maintains precise flow rates at pressures up to 1300 bar. It is ideal for applications, such as long, shallow gradients or high throughput.

[See ordering information](#)

Eliminate Metal-Induced Artifacts

PEEK-lined stainless steel columns



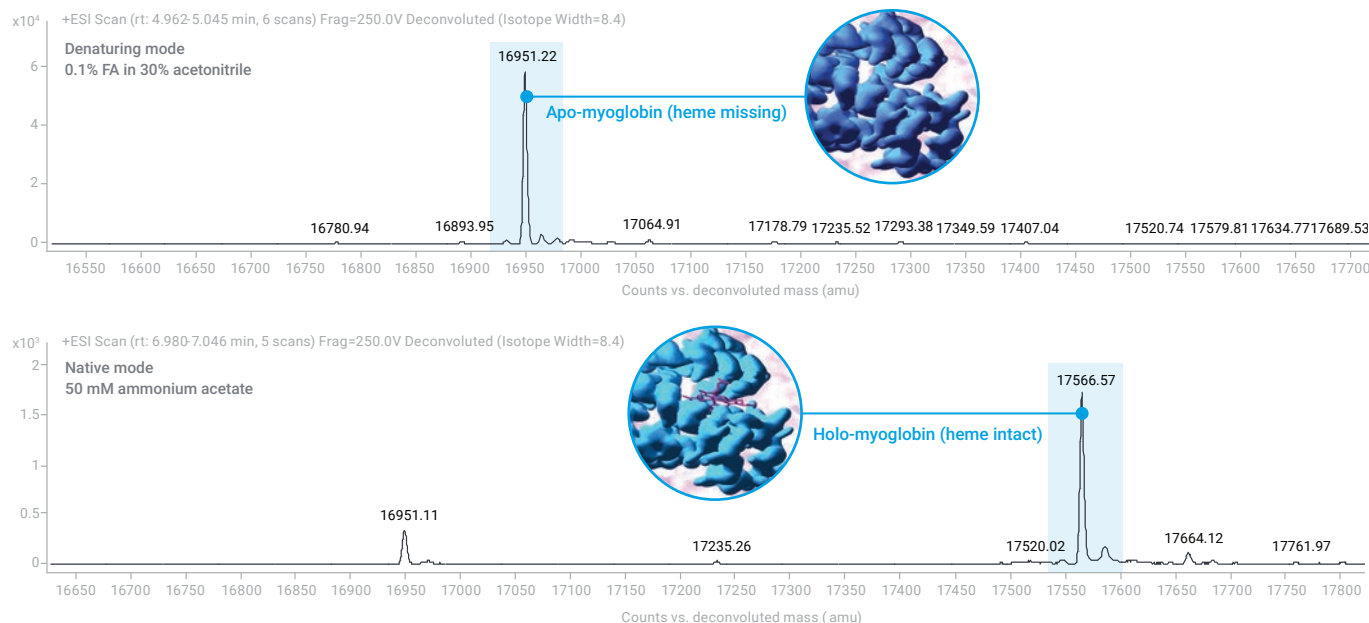
AdvanceBio SEC 1.9 μ m columns

The best choice for SEC/MS applications

These PEEK-lined columns alleviate interactions with stainless steel surfaces, which can lead to additional aggregation. They feature a narrow 2.1 mm internal diameter for operating flow rates of just 0.05 to 0.10 mL/min, and are ideal for sensitive, native-mode protein analysis. What's more, their inert hydrophilic coating allows you to use volatile mobile phases in lower concentrations while minimizing nonspecific interactions.

Because most stainless steel columns have exposed active metal sites, they require priming with biologic samples in order to obtain an acceptable level of reproducibility. PEEK-lined stainless steel columns have no active metal sites, which reduces the need for column priming—and saves you time and sample. You can also use AdvanceBio SEC standards to check these columns for quality, just as you would with stainless steel SEC columns.

PEEK-lined SEC/MS analysis of myoglobin—deconvoluted



Size exclusion chromatography of myoglobin under denaturing conditions (using formic acid as ion pair reagent) in aqueous acetonitrile on an Agilent 6545XT AdvanceBio LC/Q-TOF system. Under denaturing conditions, the deconvoluted mass corresponds to apo-myoglobin, where the heme group is no longer present. But under native mode conditions, the deconvoluted mass corresponds with the holo-myoglobin (heme group still intact), with only a small proportion of apo-myoglobin present.

[See ordering information](#)

Master your chromatography

Controlled through Agilent OpenLab software, InfinityLab LC Series instruments optimize your bio-analysis LC workflows by reducing the time you spend on data processing, review, and reporting.

Learn more about OpenLab software suite:
www.agilent.com/chem/openlab

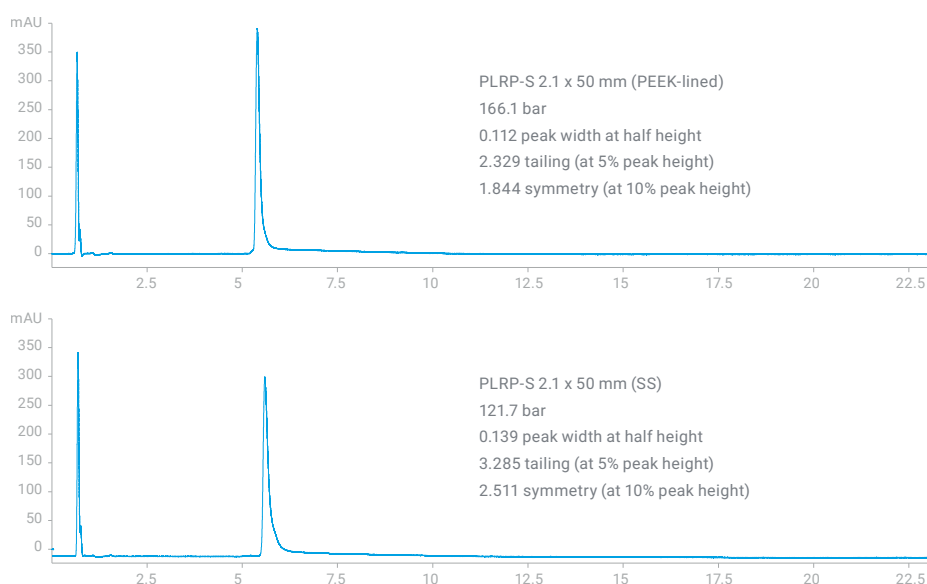
Agilent PLRP-S 1000 Å 5 µm columns

Excellent inertness for intact protein analysis

PLRP-S media is an industry leader for intact protein analysis. Available in a wide pore size for excellent mass transfer, the inert PLRP-S 1000 Å polystyrene-divinylbenzene stationary phase ensures excellent protein recovery with minimal carryover. Its polymer base confers inherent hydrophobicity for reverse-phase separations using MS-compatible ion pair reagents like formic acid. Plus, the absence of residual silanol groups allows for excellent peak shape. In addition, the column's PEEK-lined hardware minimizes the risk of artifacts—such as oxidation and peak tailing—due to secondary metal interactions.

Ramucirumab (1 mg/mL; 1 µL injection)

0.21 mL/min, 70 °C, 20–80% ACN (0.1% TFA) over 20 minutes, UV 280 nm



For intact analysis, PEEK-lined stainless steel columns can provide significant improvements in symmetry by reducing peak tailing due to secondary metal interactions while retaining similar elution profiles and retention times.



Analytical success depends upon the quality of your reference material

Agilent standards are rigorously tested and manufactured using ISO certifications—including ISO 17025 and 17034. So you can calibrate with confidence and maximize accuracy. [Learn more about Agilent NISTmAb](#)



Expand your scope with LC/MS

Whether you're analyzing large biomolecules, peptides, or glycans, the Agilent 6545XT AdvanceBio LC/Q-TOF is designed for biopharma. Together with the 1290 Infinity II bio LC and Agilent MassHunter BioConfirm software, the system automates intact protein, peptide mapping, and glycan analysis workflows.

[Learn more by downloading our brochure here](#)

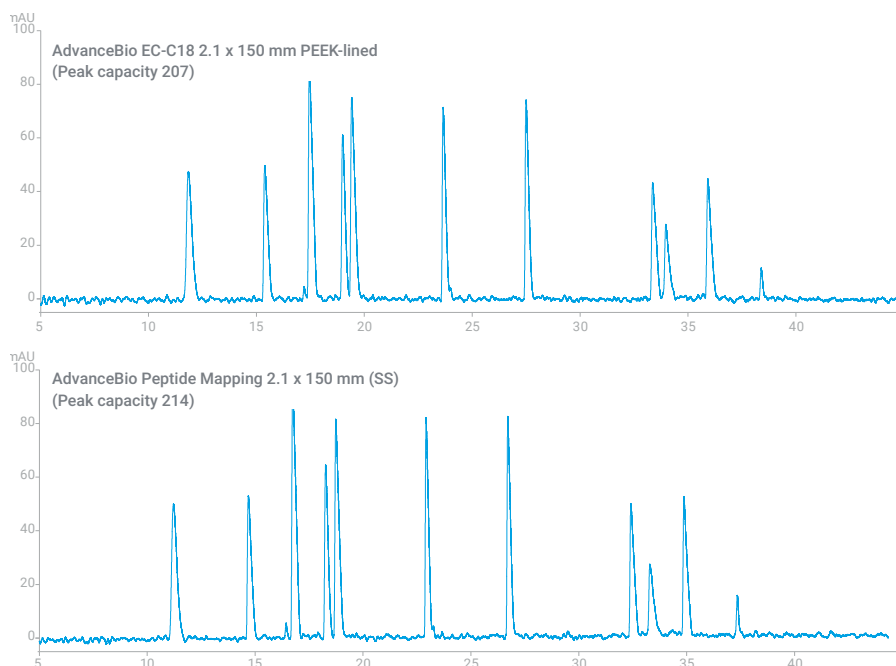
AdvanceBio 2.7 μ m EC-C18 columns

Designed for biocompatible (U)HPLC instrumentation

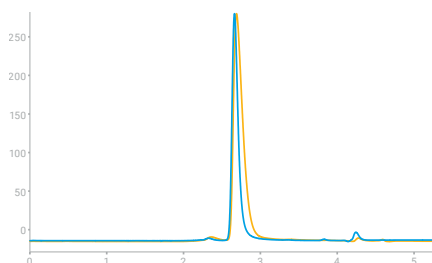
Our fully end-capped C18 silica stationary phase features excellent retention characteristics for a wide range of peptides typically found in a digested protein. What's more, their superficially porous Poroshell particles offer high performance without exceptional operating pressures. We recommend EC-C18 columns for applications where stainless steel columns might interact with components like phosphorylated peptides—or increase on-column oxidation of susceptible amino acids like methionine.

Ten peptide standard (5190-0583)

3–47% ACN (0.1% FA), 2–46 min, 0.4 mL/min; 55 °C; UV, 220 nm



For intact analysis, PEEK-lined stainless steel columns can provide significant improvements in symmetry by reducing peak tailing due to secondary metal interactions while retaining similar elution profiles and retention times.



To improve peak width and capacity, use an ultralow dispersion kit

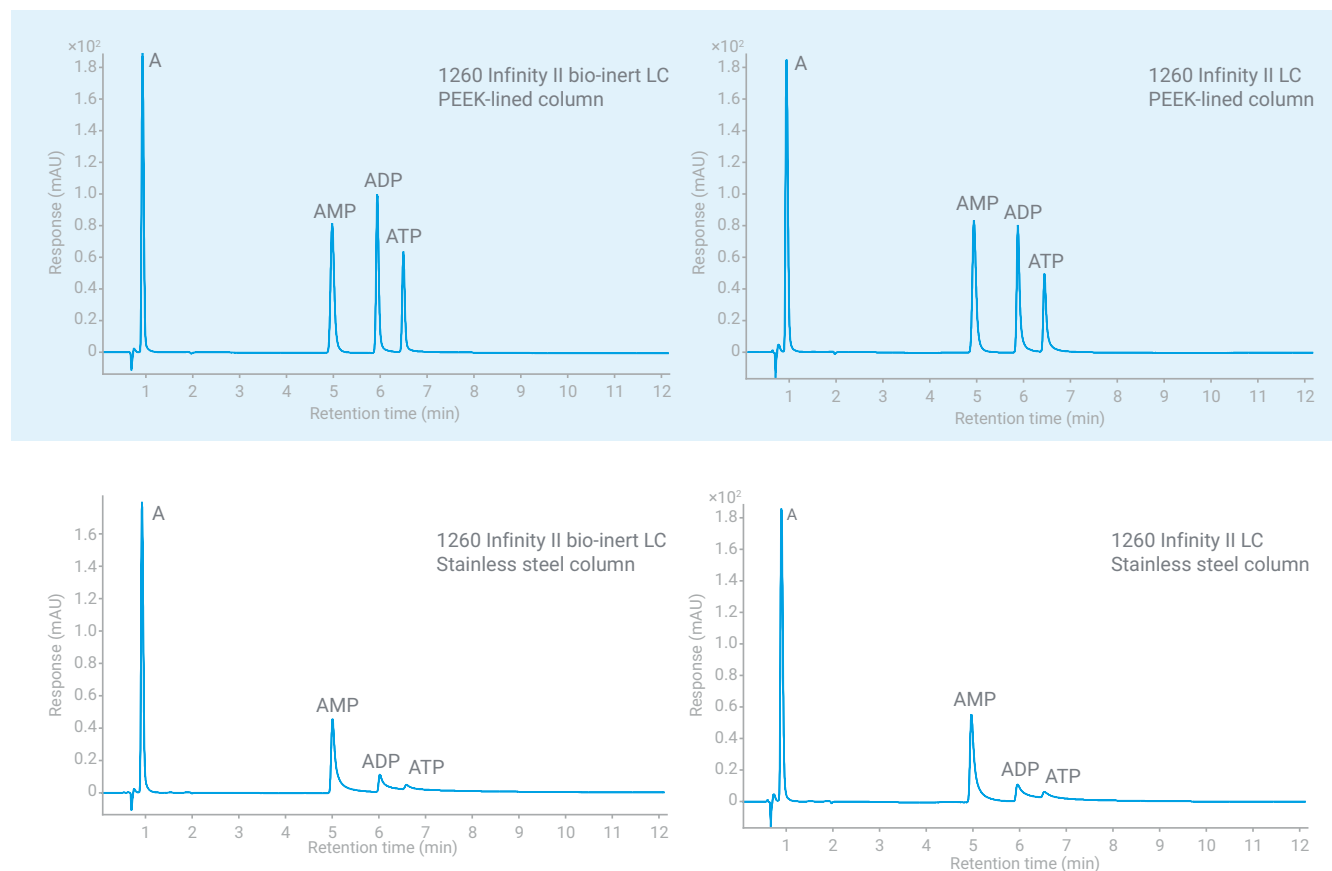
Ultralow dispersion kits allow for the least possible on-column dispersion for UHPLC methods by use of optimized tubing components. Here, the 0.07 mm ID tubing produces a much sharper peak than the 0.17 mm ID tubing. This is especially important for SEC, where low flow rates contribute to increased dispersion.

[See ordering information](#)

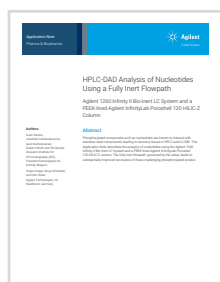
Is your flow path biocompatible?

Removing stainless steel from your flow path allows for increased recovery, improved peak shape, and higher sensitivity

As illustrated here, stainless steel column hardware (bottom) is the largest barrier to overcoming poor recoveries. PEEK-lined columns (top) maximize recovery and sensitivity while dramatically improving peak shape.



HILIC-DAD chromatograms of adenosine (A), AMP, ADP, and ATP on a stainless steel and PEEK-lined InfinityLab Poroshell 120 HILIC-Z column installed on a 1260 Infinity II bio-inert LC and 1260 Infinity II LC.



Improved recoveries of challenging phosphorylated analytes

This application was performed using InfinityLab Poroshell 120 HILIC-Z columns—both SS and PEEK-lined SS. Superficially porous particle technology is a great way to save time and solvent while increasing throughput and recovery.

Want to know more? Read the complete application note:

[HPLC-DAD Analysis of Nucleotides Using a Fully Inert Flow Path](#)

[See ordering information](#)

Ordering Information

One-click ordering



You can always count on Agilent to support your entire workflow—including sample preparation, columns, supplies, standards, and instruments. To add items to your shopping cart at the Agilent online store, simply click the part number links. Then, enter the quantities for the products you need.

Agilent columns

Description	Part Number
AdvanceBio SEC 1.9 μm 200 Å, 2.1 x 150 mm PEEK-lined	PL1980-3201PK
AdvanceBio SEC 1.9 μm 200 Å, 2.1 x 50 mm PEEK-lined	PL1980-1201PK
AdvanceBio SEC 1.9 μm 120 Å, 2.1 x 150 mm PEEK-lined	PL1980-3250PK
AdvanceBio SEC 1.9 μm 120 Å, 2.1 x 50 mm PEEK-lined	PL1980-1250PK
PLRP-S 5 μm 1000 Å, 2.1 x 100 mm PEEK-lined	PL1912-2502PK
PLRP-S 5 μm 1000 Å, 2.1 x 50 mm PEEK-lined	PL1912-1502PK
AdvanceBio EC-C18 2.7 μm , 2.1 x 100 mm PEEK-lined	675775-902
AdvanceBio EC-C18 2.7 μm , 2.1 x 50 mm PEEK-lined	679775-902
AdvanceBio MS Spent Media 100 Å, 2.1 x 100 mm	675775-901
AdvanceBio MS Spent Media 100 Å, 2.1 x 50 mm	679775-901
AdvanceBio MS Spent Media, 100 Å, 2.1 x 150 mm, 2.7 μm	673775-901



AdvanceBio PEEK-lined column
p/n PL1980-3201PK

Agilent-NISTmAb and SEC standards

Description	Part Number
Agilent NISTmAb 25 μL (QTY 1)	5191-5744
Agilent NISTmAb 25 μL (QTY 4)	5191-5745
AdvanceBio SEC 130 Å (1.5 mL)	5190-9416
AdvanceBio SEC 300 Å (1.5 mL)	5190-9417



Agilent NISTmAb standards
p/n 5191-5745

AdvanceBio Gly-X N-glycan prep with InstantPC kits (formerly ProZyme)

Description	Part Number	
	24 count	96 count
Gly-X with InstantPC kit	GX24-IPC	GX96-IPC
Gly-X with 2AB kit	GX24-2AB	GX96-2AB
AdvanceBio InstantPC Human IgG N-glycan library	GKPC-005	
AdvanceBio 2-AB Human IgG N-glycan library	GKSB-005	



AdvanceBio SEC standards
p/n 5190-9416 / 5190-9417

[Download](#) our glycan standards chart, which includes structures.

InfinityLab Quick Connect bio-inert capillaries and supplies

Description	Part Number
LC mobile phase filtration	
InfinityLab solvent filtration assembly (includes 250 mL funnel, membrane holder base, 1 L flask, and aluminum clamp)	5191-6776
InfinityLab solvent filtration 2 L glass flask (optional)	5191-6781
Regenerated cellulose filter membrane, 47 mm diameter, 0.20 µm pore size, 100/pk	5191-4340
Regenerated cellulose filter membrane, 47 mm diameter, 0.45 µm pore size, 100/pk	5191-4337
Column connection	
InfinityLab Quick Connect LC fitting	5067-5965
Quick Connect Capillary MP35N 0.12 x 105 mm	5500-1578
Quick Connect Capillary MP35N 0.12 x 150 mm	5500-1579
Quick Connect Capillary MP35N 0.12 x 220 mm	5500-1580
Quick Connect Capillary MP35N 0.12 x 280 mm	5500-1581
Quick Connect Capillary MP35N 0.12 x 400 mm	5500-1582
Quick Connect Capillary MP35N 0.12 x 500 mm	5500-1583
Quick Connect Capillary MP35N 0.17 x 105 mm	5500-1584
Quick Connect Capillary MP35N 0.17 x 150 mm	5500-1585
Quick Connect Capillary MP35N 0.17 x 220 mm	5500-1586
Quick Connect Capillary MP35N 0.17 x 280 mm	5500-1587
Quick Connect Capillary MP35N 0.17 x 500 mm	5500-1588
Well plates	
Well plate 96/0.5 mL, round, U shape, 14 mm, PP, 120/pk	5043-9311
Well plate 96/1.0 mL, round, U shape, 32 mm, PP, 50/pk	5043-9305
Mat 96, round, preslitted, silicone, 50/pk	5042-1389
Detector bio flow cells	
Max-light cartridge cell LSS 10 mm	G7117-60020
Bio micro flow cell VWD, 3 mm, 2 µL, RFID	G1314-60189
Bio standard flow cell VWD, 10 mm, 14 µL, RFID	G1314-60188



InfinityLab Quick Connect LC fitting
p/n 5067-5965



The Gly-X N-glycan rapid release and labeling with InstantPC kit uses an in-solution enzymatic protein deglycosylation, followed by rapid labeling of released N-glycans with InstantPC dye. After a simple clean-up step, the glycan samples are ready for analysis by UHPLC, LC/MS, MS/MS, and other methods. [Learn more about AdvanceBio Gly-X technology](#)

Agilent CrossLab services

CrossLab is an Agilent capability that integrates services and consumables to support workflow success and important outcomes like improved productivity and operational efficiency. Through CrossLab, Agilent strives to provide insight in every interaction to help you achieve your goals. CrossLab offers method optimization, flexible service plans, and training for all skill levels. We have many other products and services to help you manage your instruments and your lab for best performance.

Learn more about Agilent CrossLab, and see examples of insight that leads to great outcomes, at www.agilent.com/crosslab



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