

HPLC columns

Connected chromatography solutions

Low-flow columns and accessories

Introduction

Low-flow chromatography is ideal when detailed sample information is required from small sample volumes, such as proteomic, metabolomic, and intact protein analysis. The Thermo Scientific range of nano-, capillary-, and micro-flow columns offer excellent sensitivity and resolution in easy-to-use formats.

- Thermo Scientific[™] µPAC[™] Neo HPLC Columns
- Thermo Scientific[™] EASY-Spray[™] HPLC Columns
- Thermo Scientific[™] Double nanoViper[™] HPLC Columns





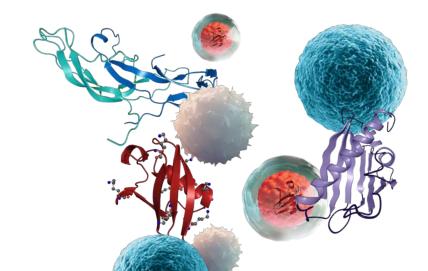




Video: Low-flow HPLC columns connectivity

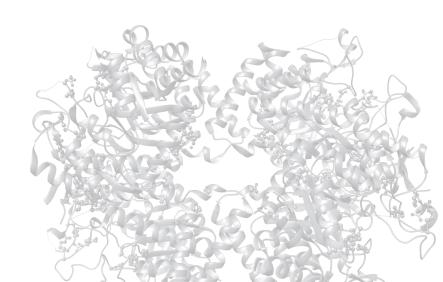
Contents

| μPAC Neo HPLC columns | 5 |
|-------------------------------|----|
| EASY-Spray HPLC columns | 8 |
| Bottom-up proteomics | 9 |
| Top-down proteomics | 10 |
| Double nanoViper HPLC columns | 12 |
| Bottom-up proteomics | 13 |
| Top-down proteomics | 14 |



Column selection guide

| | Pillar array column format | olumn format | |
|-------------------------------|---|---|---|
| | Thermo Scientific™ µPAC™ Neo HPLC Columns | Thermo Scientific™ EASY-Spray™ HPLC Columns | Thermo Scientific™ Double nanoViper™ HPLC Columns |
| Technology | February and St. 10 years (February) | 0 | -2 |
| Benefits | Ultimate separation Excellent retention time stability A unique combination of performance and reliability to get the highest sample coverage every time Separate emitters Compatible with all low-flow U/HPLC instruments | Ease-of-use • Click-and-spray connect with Thermo Scientific™ EASY-Spray™ Source • Thermo Scientific™ nanoViper™ connections • Integrated column and emitter • Integrated temperature control • For use with Thermo Scientific™ mass spectrometry systems | Analytical flexibility Universal Thermo Scientific™ nanoViper™ Fingertight Fittings for column inlet and outlet Simple zero-dead-volume (ZDV) connections Separate emitters Compatible with all low-flow U/HPLC instruments |
| Application areas/chemistries | Deliver excellent column-to-column reproducibility with flow rate flexibility. Ideally suited for proteomic analyses of HPLC separations up to 450 bar. • 50 cm column: 15–60 min gradient time • 110 cm column: 90–150 min gradient time • 50 cm low-load single cell analysis: 15–60 min gradient time | Bottom-up proteomic applications The Thermo Scientific™ PepMap™ Neo Uh our portfolio. PepMap Neo columns are pa 1500 bar pressure rating, improved colum increased efficiency. Top- and middle-down proteomic app The Thermo Scientific™ MAbPac™ Capilla best suited for the characterization of inta proteomic applications where sample am | lications ry Reversed-Phase HPLC Column is act proteins in top- and middle-down |



μPAC Neo HPLC columns



The μ PAC Neo columns are specifically suited for bottom-up proteomic applications where separation performance is critical to the success of the analysis. Our μ PAC Neo HPLC columns offer highest resolution and peak capacities for complex biological samples. The unique μ -pillar backbone improves column-to-column reproducibility and robustness, providing more confidence in analytical results.

Additional reading

| Links | Туре | Description | |
|------------|--|--|--|
| • | Reference guide | Chromatography consumables reference guide for low-flow LC-MS proteomic research | |
| • | Flyer | Enabling high sensitivity LC-MS analysis for bottom-up and top-down proteomic research | |
| (A) | Learn more thermofisher.com/lowflowHPLCcolumns | | |

Choose a µPAC Neo HPLC column when:

- · Highest resolution and peak capacities is required
- Your samples span a wide concentration range
- Highest LC-MS sensitivity is needed
- You want to speed up your runtimes
- LC-MS robustness is needed
- You want an increased column lifetime
- You prefer working at much lower back pressures than with packed bed columns
- It is important to compare results from experiments spanning over time or geographical location

What makes µPAC Neo HPLC columns special?

The unique separation path provides:

- µ-pillar stationary backbone, micromachined in a silicon wafer
- Flow path designed for highest analyte concentration during elution
- Extra high-resolution separations, using up to 110 cm column lengths
- Low back pressure separations, improving column and emitter robustness
- Perfect match with single cell proteomics sample amounts

thermo scientific

50 cm µPAC Neo

μPAC Neo HPLC columns continued

Velocity Label-free Quantitation (LFQ) Data Independent Acquisition (DIA) Platform

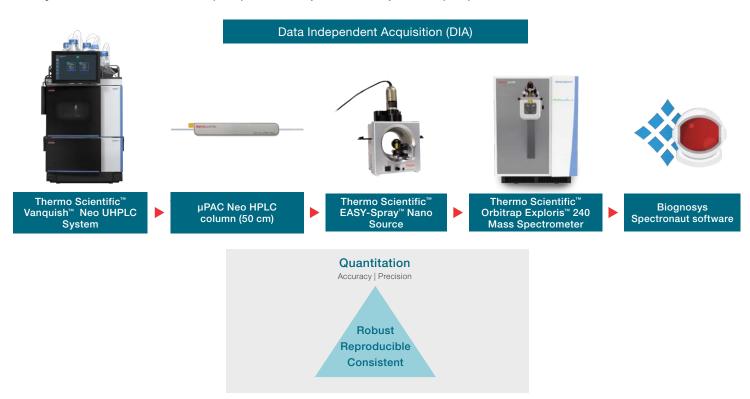


Figure 1. Graphical schematic of HR-DIA workflow for label-free quantitation of two- and three-proteome mixtures. The different components of the workflow are depicted on the top. The main goal of the setup is the quantitative performance at high sample throughput while delivering robust and reproducible results to make it a perfect fit for large scale clinical and biomarker discovery studies.

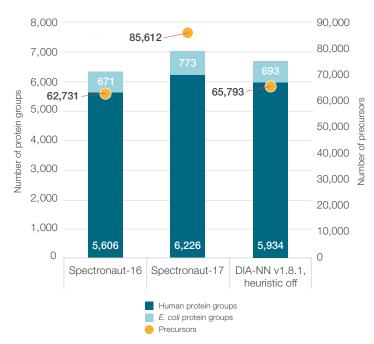


Figure 2. HR-DIA Workflow delivers confident proteome coverage utilizing next generation library-free analysis approaches. Bar graph comparison of protein group (human and E. coli) and precursor (total) numbers identified in 12 runs of two-proteome mix by use of three different software packages. Data analysis has been done by library-free analysis. All protein group results are filtered for 1% experiment-wide FDR.

μPAC Neo HPLC columns continued





Ordering information

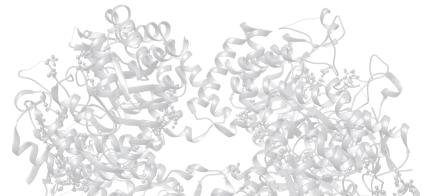
| Description | Pillar dimensions (µm) | Interpillar distance (µm) | Column length (cm) | Flowrate range (nL/min) | Cat. no |
|---|------------------------------|---------------------------------|-----------------------|-------------------------------|-----------------|
| Thermo Scientific 50 cm µPAC Neo column | 2.5 | 1.25 | 50 | 100-750 | COL-nano050NeoB |
| Thermo Scientific 110 cm µPAC Neo column | 2.5 | 1.25 | 110 | 100-750 | COL-nano110NeoB |
| Thermo Scientific 50 cm µPAC Neo low-load column | 2.5 | 1.25 | 50 | 100-750 | COL-IoloNeoB |

Ordering information

| Description | Pillar dimensions (µm) | Interpillar distance (µm) | Column length (cm) | Cat. no |
|---|------------------------------|---------------------------------|-----------------------|------------------|
| Thermo Scientific™ µPAC™ Neo Trapping Column | 5 | 2.5 | 1 | COL-trploloNeoB2 |

Ordering information

| Description | Pillar dimensions (µm) | Details | For use with | Cat. no |
|---|------------------------------|---|--------------------------|---------|
| Thermo Scientific™ EASY-Spray™ Nano Emitters | 10 | Bullet type without transfer line | EASY-Spray ion-source | ES993 |



EASY-Spray HPLC columns



Ensure robust nano- and capillary-flow LC-MS analysis using Thermo Scientific EASY-Spray HPLC Columns. The integrated column/emitter design eliminates dead volume and is temperature-controlled for maximum reliability and performance. Rigorously tested to ensure maximum quality, these columns deliver maximum simplicity and ease-of-use. The capillary-flow HPLC columns provide sensitive protein, peptide, and monoclonal antibody (MAb) separation. They give proteomic researchers more than ever before: more throughput, more sensitivity, more separation power, and more ease of use.

Additional reading

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| (A) | Learn more thermofisher.com/lowflowHPLCcolumns | | |

Choose an EASY-Spray column when:

- You want simple connections with an EASY-Spray source.
 This is ideal for novice and experienced users
- · Sample amount is limited
- Analytical UHPLC does not provide sufficient sensitivity
- Workflow simplicity is key
- High sensitivity is required to identify proteins and peptides at low expression levels
- Analyses are done in a targeted and untargeted way for screening and verification

What makes an EASY-Spray column special?

Unique design provides uncompromised performance in an ease-of-use format for nano and capillary LC-MS analysis

Features for optimum data quality:

- Simple connection to the LC and Thermo Scientific MS instruments
- Precision machined and positioned glass emitters
- Integrated nanoViper zero-dead-volume (ZDV) unions
- Integrated temperature control



Video:
Thermo Scientific EASY-Spray
150 mm LC columns



EASY-Spray HPLC columns continued

Bottom-up proteomics



PepMap Neo HPLC columns

The Thermo Scientific™ EASY-Spray™ PepMap™ Neo UHPLC Columns are perfect for bottom-up proteomic applications. Packed at higher pressure and rated to 1500 bar, they provide consistent column-to-column performance, long column lifetime, and excellent efficiency. These benefits are true at any pressure.

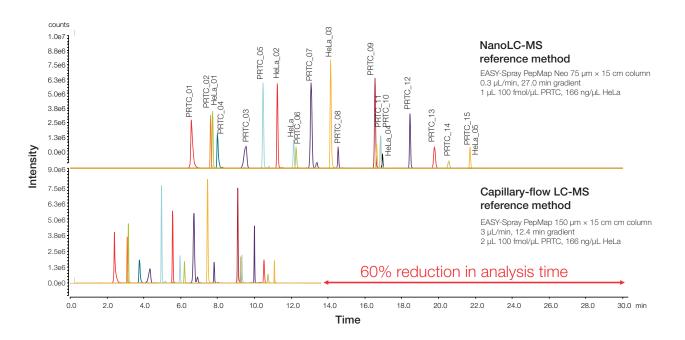


Figure 3. The 60% reduction in total analysis time allows increasing the sample throughput moving from the nanoto the capillary-flow LC-MS method



Ordering information for bottom-up proteomic applications

| Description | Length (mm) | Column ID (µm) | Cat. no |
|-------------------------------------|-------------|----------------|------------------|
| | 150 | 75 | ES75150PN |
| EASY-Spray PepMap Neo UHPLC Columns | 500 | 75 | ES75500PN |
| | 750 | 75 | <u>ES75750PN</u> |



EASY-Spray HPLC columns continued

Top-down proteomics



MAbPac Capillary Reversed Phase HPLC Column

The Thermo Scientific™ MAbPac™ Capillary Reversed Phase capillary column is best suited for the characterization of intact proteins in top-down proteomic, clinical, and anti-doping applications where sample amount is limited or sensitivity is crucial.

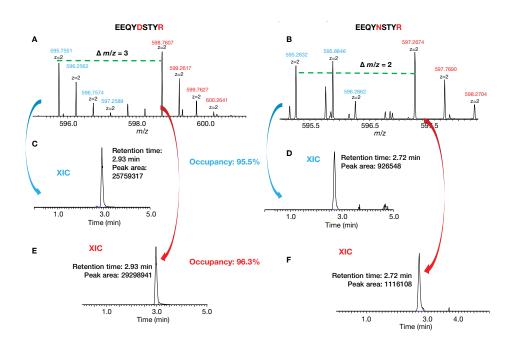


Figure 4. Calculation of site occupancy of N306 in Fab glycosylated mAb



Ordering information for top-down proteomic applications

| Description | Length (mm) | Column ID (µm) | Cat. no |
|------------------------|-------------|----------------|---------|
| EASY-Spray HPLC Column | 150 | 150 | ES907 |



EASY-Spray HPLC columns continued



EASY-Spray accessories

For the best performance from your EASY-Spray column consider investing in these accessories.

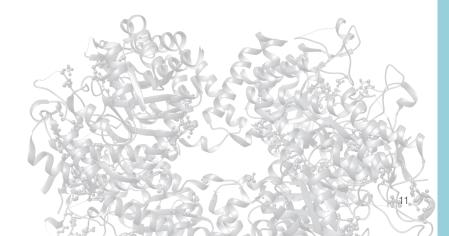


Ordering information

| Description | Union type | Particle size (µm) | Column ID (µm) | Media bed length (mm) | Trap length (mm) | Cat. no |
|--|------------------|-----------------------|-------------------|-----------------------------|------------------------|---------------|
| Thermo Scientific™ PepMap™ Neo Trap Cartridge | N/A | 5 | 300 | 5 | N/A | <u>174500</u> |
| | Nut/sleeve | 5 | 200 | 20 | 150 | <u>164213</u> |
| Thermo Scientific™ Acclaim™ | Nut/sleeve | 5 | 100 | 20 | 150 | <u>164199</u> |
| PepMap™ 100 C18 HPLC Column, Trap Column | Double nanoViper | 5 | 100 | 20 | 150 | 164750 |
| | Double nanoViper | 3 | 75 | 20 | 150 | <u>164535</u> |
| | Double nanoViper | 3 | 75 | 20 | 70 | 164946 |

Ordering information

| Description | For use with | Cat. no |
|---|-------------------------|---------------|
| Thermo Scientific [™] PepMap [™] Neo Trap Cartridge Holder, PEEK Tubing, and nanoViper [™] Fittings | Low-flow PepMap columns | <u>174502</u> |



Double nanoViper columns



The Thermo Scientific™ Viper™ and Thermo Scientific™ nanoViper™ Fingertight Fitting Systems provide tool-free connections designed to be used for the entire fluidic pathway in LC systems to improve chromatographic results.

Virtually without any dead-volume, Viper and nanoViper fittings combine usability with high performance. Viper and nanoViper connections can be used on all standard LC modules, valves, and columns quickly, independent of different connection geometries and system backpressures. Dedicated capillary kits for standard LC system configurations and application-specific setups enable high qualitative and reproducible results for all flow rates and pressure ranges.

Additional reading

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|------------|--|--|--|
| • | Reference guide | Chromatography consumables reference guide for low-flow LC-MS proteomic research | |
| • | Flyer | Enabling high sensitivity LC-MS analysis for bottom-up and top-down proteomic research | |
| • | Product specifications | Viper and nanoViper Fingertight Fitting Systems | |
| (A) | Learn more thermofisher.com/lowflowHPLCcolumns | | |

Choose these columns when:

- Maximum flexibility is required
- Changing the emitter and column independently is important





What makes these columns special?

These stand-alone nano-, capillary-, and micro-flow columns are:

- Designed with single nanoViper and double nanoViper fingertight fittings for trouble-free connection
- For robust separation in proteomics research, drug discovery, and high-throughput proteomics laboratories



Video:

Discover a better LC connection

Double nanoViper columns continued

Bottom-up proteomics



Double nanoViper PepMap Neo UHPLC columns

Separate challenging peptide mapping samples with Thermo Scientific™ Double nanoViper™ PepMap™ Neo UHPLC Columns. These columns feature easy connectivity, high reproducibilty, and excellent separations. Our Neo columns are packed to higher pressure and provide 1500 bar pressure capability, improved column-to-column consistency, and increased efficiency. The column media is manufactured and selected to exacting standards and packed at high pressure, resulting in enhanced peak symmetry, resolution, and column-to-column reproducibility that allows you to obtain greater sample coverage and sample insights.

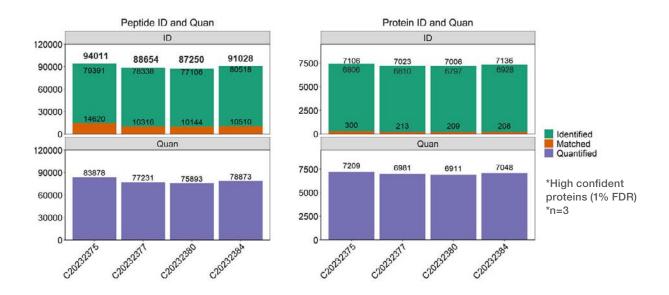


Figure 5. Reproducible identification and quantification of HeLa peptides and proteins over 4 EASY-Spray PepMap Neo columns while using the Vanquish Neo UHPLC system coupled with the Orbitrap Exploris 480 mass spectrometer



Ordering information for bottom-up proteomic applications

| Format | Length (mm) | Column ID (µm) | Cat. no |
|--|-------------|----------------|-------------------|
| Double nanoViper PepMap Neo UHPLC Columns | 150 | 75 | <u>DNV75150PN</u> |
| | 500 | 75 | <u>DNV75500PN</u> |
| | 750 | 75 | <u>DNV75750PN</u> |



Double nanoViper columns continued

Top-down proteomics



MAbPac Capillary Reversed Phase HPLC Column

The Thermo Scientific MAbPac Capillary Reversed Phase column is best suited for the characterization of intact proteins in top-down proteomic, clinical, and anti-doping applications where sample amount is limited or sensitivity is crucial.



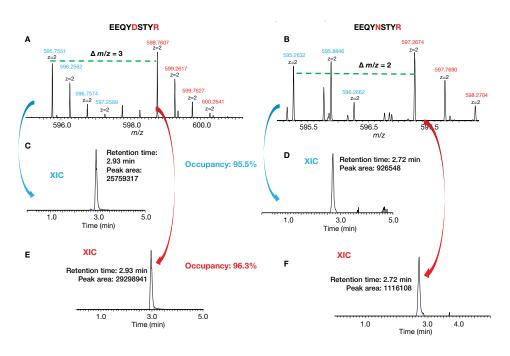


Figure 6. Calculation of site occupancy of N306 in Fab glycosylated mAb



Ordering information for top-down proteomic applications

| Format | Length (mm) | Column ID (µm) | Cat. no. | |
|--|-------------|----------------|---------------|--|
| MAbPac Capillary Reversed Phase HPLC Column | 150 | 150 | <u>164947</u> | |



Double nanoViper columns continued



LC-MS connection accessories and emitters

These emitters, nanoViper tubing kits, and unions offer easy connection from your LC system to an EASY-Spray source.



Ordering information

| Description | For use with | Part number | |
|--|----------------------------|-------------|--|
| Viper and nanoViper Fingertight Fittings Accessories | | 6040.2304 | |
| nanoViper Fingertight Fittings, 20 μm x 550 mm | Double papel/inex celumns | 6041.5260 | |
| EASY-Spray Nano Emitter, 10 μm | - Double nanoViper columns | ES993 | |
| EASY-Spray Capillary Emitter, 15 μm | _ | ES994 | |

Traps and accessories

For the best performance from your double nanoViper column consider investing in these nanotraps.



Ordering information

| Description | Union type | Particle size (µm) | Column ID (µm) | Media bed length (mm) | Trap length (mm) | Cat. no |
|--|------------------|-----------------------|-------------------|-----------------------------|------------------------|---------------|
| Thermo Scientific™ PepMap™ Neo Trap Cartridge | N/A | 5 | 300 | 5 | N/A | <u>174500</u> |
| Thermo Scientific™ Acclaim™ PepMap™ 100 C18 HPLC Column, Trap Column | Nut/sleeve | 5 | 200 | 20 | 150 | <u>164213</u> |
| | Nut/sleeve | 5 | 100 | 20 | 150 | 164199 |
| | Double nanoViper | 5 | 100 | 20 | 150 | 164750 |
| | Double nanoViper | 3 | 75 | 20 | 150 | <u>164535</u> |
| | Double nanoViper | 3 | 75 | 20 | 70 | 164946 |