µPAC Neo HPLC columns
Use and care instructions

Product:
Thermo Scientific™ µPAC™ Neo Low-load Trapping Column

Pressure limit
Do not exceed the maximum trapping column pressure of 400 bar (5,800 psi). Exceeding this value will cause irreversible damage to the trapping column.

Installing the trapping column
The µPAC Neo low-load trapping column is perfectly bi-directional, with integrated Thermo Scientific™ nanoViper™ Fingertight Fittings on both connection tubings. It can be used in either direction without the risk of damaging the separation bed.

Recommended configuration
The µPAC Neo low-load trapping column is a C8 chemistry trapping column and designed to work in combination with the Thermo Scientific™ µPAC™ Neo Low-load Column, for analysis of samples ≤10 ng of digested protein. The trapping column can be used in both forward flush and backflush mode, depending on the user’s preference and the nanoLC system’s capabilities.

Backflush trapping
The µPAC Neo low-load trapping column is connected directly to the UHPLC switching valve, either to the autosampler valve in the Thermo Scientific™ Vanquish™ Neo Autosampler or to the column oven valve.

Vented-tee
The µPAC Neo low-load trapping column is configured in-line with the µPAC Neo low-load column via an internal reducing connector T-piece.

On the Thermo Scientific™ Vanquish™ Neo UHPLC System, we would recommend the backflush configuration.

Trapping column operation
Internal volume
The µPAC Neo low-load trapping column has an internal volume of 900 nL.

Column pressure
Maximum operating pressure is 400 bar (5,800 psi).

Flow rate
The µPAC Neo low-load trapping column can be operated at flow rates between 1.0 to 60 µL/min. At 10 µL/min, using a water-acetonitrile mobile phase solvent system, the back pressure over the µPAC Neo low-load trapping column ranges from 15 to 30 bar.

Column temperature
Maximum operating temperature is 60 °C (140 °F).

pH range
2.0 < pH < 7.0.
Trapping column operation (continued)

Table 1. Properties overview

<table>
<thead>
<tr>
<th>Column</th>
<th>Stationary phase</th>
<th>Max. column pressure</th>
<th>Max. temp (°C)</th>
<th>pH stability</th>
<th>Column volume (µL)</th>
<th>Loadability (µg protein)</th>
<th>Flow rate (µL/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>µPAC Neo low-load trapping column</td>
<td>C8</td>
<td>400 bar</td>
<td>60</td>
<td>2-7</td>
<td>900</td>
<td>&lt;0.01</td>
<td>0.1-60</td>
</tr>
</tbody>
</table>

Typical solvents
Acetonitrile (ACN), methanol (MeOH), isopropanol (IPA), trifluoroacetic acid (TFA), formic acid (FA)

Sample solvent
To maximize trapping efficiency, we recommend to dissolve samples in 0.1% TFA containing a low percentage of organic modifier (e.g. 1% acetonitrile). For backflush trap-elute configurations, a loading buffer comprised of 1% acetonitrile, 0.1% TFA is strongly advised.

Sample capacity
For the analysis of tryptic digested proteins, the equivalent of 10 ng of total protein can be injected without overloading the µPAC Neo low-load trapping column:
- Use only fresh, degassed and LC-MS grade mobile phases that are compatible with reversed-phased liquid chromatography (RP-LC)
- Switch only between mutually miscible mobile phases
- Cleanliness of the sample greatly affects column and trapping column life
- Use (clean) sample which are free from particulates. Filter if necessary, using a 0.5 µm cut-off filter.

Disclaimer and contact
Warranty of the column extends up to 30 days after the purchase of the product.