YMC User Manual
Care and Use Instructions

YMC Capillary Columns

1. Introduction

Thank you for purchasing a YMC capillary column. They are compatible with most Nano-/MicroLC/MS systems. Capillary columns are suitable for extremely low sample volumes and low flow rates. They are available either with 1/16" connections (10–32 thread) or with 1/32" connections (6–40 thread).

2. Specifications

For selected phases*

<table>
<tr><th>Packing material</th><th>Particle size (µm)</th><th>Pore Size (nm)</th><th>Usable pH range**</th><th>Max. Temp. (°C)</th><th>Max. Pressure</th></tr>
<tr><td>YMC-Triart C18</td><td rowspan="8">1.9; 3; 5</td><td>12</td><td>1–12</td><td rowspan="5">pH<7: 90
pH>7: 50</td><td rowspan="16">1.9 µm:
600 bar / 8,700 psi
for 300 / 500 µm ID

2; 3; 5 µm:
550 bar / 7,975 psi
for 300 / 500 µm ID

2; 3; 5 µm:
450 bar / 6,525 psi
for 75 / 100 µm ID</td></tr>
<tr><td>YMC-Triart C18 ExRS</td><td>8</td><td>1–12</td></tr>
<tr><td>YMC-Triart Bio C18</td><td>30</td><td>1–12</td></tr>
<tr><td>YMC-Triart C8</td><td>12</td><td>1–12</td></tr>
<tr><td>YMC-Triart Bio C4</td><td>30</td><td>1–10</td></tr>
<tr><td>YMC-Triart Diol-HILIC</td><td>12</td><td>2–10</td><td rowspan="8">50</td></tr>
<tr><td>YMC-Triart PFP</td><td>12</td><td>1–8</td></tr>
<tr><td>YMC-Pack Pro C18</td><td>2; 3; 5</td><td>12</td><td>2–8</td></tr>
<tr><td>Hydrosphere C18</td><td>2; 3; 5</td><td>12</td><td>2–8</td></tr>
<tr><td>YMC-Pack Pro C18 RS</td><td>3; 5</td><td>8</td><td>1–10</td></tr>
<tr><td>YMC-Pack Pro C8</td><td rowspan="4">3;5</td><td>12</td><td rowspan="3">2–7.5</td></tr>
<tr><td>YMC-Pack Pro C4</td><td>12</td></tr>
<tr><td>YMC-Pack ODS-A</td><td>12; 20; 30</td></tr>
<tr><td>YMC-Pack ODS-AQ</td><td>12; 20</td></tr>
<tr><td>YMC-Pack Diol-60/-120 SEC</td><td>6; 12</td><td rowspan="3">5–7.5</td><td rowspan="3">40</td></tr>
<tr><td>YMC-Pack Diol-200/-300 SEC</td><td>2; 3; 5</td><td>20; 30</td></tr>
<tr><td>YMC-SEC MAB</td><td>3</td><td>25</td></tr>
</table>

* Refer to the Care & Use Instructions for the corresponding product/brand if yours is not listed above.

** pH range of the phases. Due to the glass lining of the column we recommend to use pH values with an upper limit of 10 unless the phase demands a lower limit.

Typical flow rates for capillary columns

Column ID	Typical flow rates for RP *
0.5 mm	10–30 µL/min
0.3 mm	3–10 µL/min
150 µm	0.7–1.1 µL/min
100 µm	300–500 nL/min
75 µm	100–300 nl/min

* Depending on length and particle size.
In HILIC mode flow rates are recommended to be reduced by half.

3. Shipping Solvent

Indicated in the COLUMN INSPECTION REPORT.
Replace with this solvent for storage.

4. Mobile Phase

[Common instruction]

- The correct direction of the solvent flow is indicated by an arrow on the column identification label.
- Recommendations of pH for column use are shown in the specifications table in section 2. When using the column near the upper or lower limit, a mobile phase containing 10 % concentration of organic solvent should be used. The column lifetime will be reduced under certain conditions by temperature and mobile phase composition.
- Mobile phase and sample should be filtered before analysis to achieve reproducible results and long column lifetime.

[Reversed-phase columns]

- Aqueous or non-aqueous solvent can be used as a mobile phase for reversed-phase HPLC. Repetitive switching between solvents with large difference in polarities might degrade the column performance. In general, acetonitrile, methanol and tetrahydrofuran (THF) are recommended for regular use. When using THF as a mobile phase, be aware that the solvent resistance within your system or tubing (PEEK parts are especially unsuitable for use with THF)
- When using YMC-Triart C18 ExRS or YMC-Pack Pro C18 RS, the extremely high hydrophobicity of the phase might cause difficulties with replacement or equilibration of mobile phases containing low concentration of organic solvent. The mobile phase should contain more than 15 % of methanol or 10 % of other lower polar organic solvent. When replacing aqueous/methanol solutions with aqueous/acetonitrile solutions, mobile phases containing less than 20 % acetonitrile may result in irregularities in retention time or peak shapes. In this case, first replace with an aqueous solution containing 60 % acetonitrile, and then replace with the mobile phase.

[YMC-Triart Diol-HILIC]

- The most suitable mobile phase is acetonitrile/water or buffer (approx. 90/10 – 60/40). In addition, general water-miscible organic solvents can be used. Contrary to reversed phase separations, with HILIC separations, the lower polarity of mobile phase should contain at least 3 % of aqueous solution to enhance the separation reproducibility by forming a stable hydrated layer on the surface of packing material. Ammonium acetate or ammonium formate are the most recommended buffer salts. Usually, 10–20 mM final salt concentration in a mobile phase is sufficient; adjustment in the range of 5–200 mM can be made. In case of gradient elution, each mobile phase should be adjusted to maintain a constant salt concentration during each separation. Before using or replacing a mobile phase, please confirm precipitation of salts is not observed. Avoid phosphate salts and other low solubility buffers with organic solvents. Where possible dissolve the sample in a solvent that is of the same composition as the initial mobile phase. Consider the miscibility of the sample with the mobile phase to avoid precipitation of solutes or salts contained in the sample.

[Size exclusion columns]

- Aqueous mobile phases are normally used. Total salt concentration of the mobile phase should be less than 0.7 M. Phosphate, Tris-HCl, citrate, etc. are all suitable as buffer solutions. Also, aqueous solutions of urea and guanidine hydrochloride which are used as a denaturant of proteins can be used. Moreover, 0.1 % or less concentration of surface-active agents such as Tween80 or SDS are also suitable. Alcohol or acetonitrile can be added to mobile phase. Be aware of any rise in operating pressure due to the possibility of precipitation of buffer salts/additives when using a mobile phase containing alcohol or acetonitrile.

5. Column Cleaning

[Reversed-phase columns]

- Flush the column with a solution containing a higher ratio of organic solvent to remove any compounds that have a great capacity for retention in the column after using mobile phases not containing buffer salts/additives. Concentrations of organic solvents up to 100 % can be used. A cleaning solution containing THF might be effective when removing highly hydrophobic (lipid-soluble) substances that are absorbed onto the gel.
- When using mobile phases containing buffer salts/additives, first replace this with water/organic solution containing no buffer salts/additives (A ratio of water to organic solvent should be set at the same proportions as a mobile phase). Then flush the column in accordance with the method described above. Mobile phases containing about 50 mM or less of buffer salts/additives can be replaced directly with 60 % acetonitrile aqueous solution.
- Flushing with 100 % water after using the column around the pH limit might shorten the column lifetime. Flushing the column with a water/organic solution as described above, such as 60 % acetonitrile aqueous solution is recommended.
- Once macromolecules such as proteins or polysaccharides are absorbed onto the gel, they are very difficult to remove, even if solvents with high eluting capability are used. To avoid contamination of the column with such compounds, conduct sample pre-treatment carefully before introduction into the column.

[YMC-Triart Diol-HILIC]

- Use a mixture of organic solvent and water with a higher solvent strength than the mobile phase, such as acetonitrile/water (50/50), to remove strongly retained substances. Usually, a solvent of 50 % water is sufficient to remove polar contaminants. If further cleaning is required, flush the column with acetonitrile/water (5/95).

[Size exclusiom columns]

- Flush the column with solvent containing high salt concentration (approx. 0.5 M), if some hydrophobic proteins or materials are absorbed or retained. Be careful not to exceed the usable pH.

6. Accessories

A column coupler is supplied with every pack of capillary guard cartridges to guarantee the optimum connection with low dead volume. A stainless steel coupler is provided for 1/32" and 1/16" columns. Every coupler can be purchased separately if required.

P/N	Description	Units
XRCP3201E-1PK	1/32" OD, 0.13 mm ID, 6-40	1
XRCP3201E-3PK	1/32" OD, 0.13 mm ID, 6-40	3
XRCP3201E-5PK	1/32" OD, 0.13 mm ID, 6-40	5
XRCP1601E-1PK	1/16" OD, 0.1 mm ID, 10-32	1

For use with Eksigent Micro- and NanoLC systems, order columns with 1/32" (6–40 thread) end-fittings and use either, Eksigent 6/40 fitting p/n 5019621 or VALCO p/n ZNF.5FPK.

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