GE Healthcare
Life Sciences

Instructions 71-7146-00 AI **Ion Exchange Columns**

RESOURCE™ Q, 1 ml and 6 ml
RESOURCE S, 1 ml and 6 ml

Introduction

RESOURCE Q and S are pre-packed columns for separating biomolecules by ion exchange chromatography. The columns are pre-packed with SOURCE™ 15Q or SOURCE 15S. The medium is based on rigid, monodisperse 15 µm beads made of polystyrene/divinyl benzene. The small monodisperse bead give high-resolution purification at high flow rates. In addition, hydrophilisation of the beads minimizes non-specific adsorption and allows high recovery of purified sample. The material of the column body is PEEK (polyetheretherketone). The top frit is made of titanium. The bottom filter is made of polypropylene. They connect to ÄKTA™ design systems or other chromatography systems.

Intended use

The RESOURCE Q and S columns are intended for research use only, and shall not be used in any clinical or *in vitro* procedures for diagnostic purposes.

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Table 1. Characteristics of RESOURCE Q and S columns

Matrix	Polystyrene/divinylbenzen
Ligand	Q: Quaternary ammonium (strong anion exchanger) S: Methyl sulfonate (strong cation exchanger)
Particle size	15 µm
pH stability	2 to 12 (long term), 1 to 14 (short term)
RESOURCE prepacked columns	
Bed volume	1 ml or 6 ml
Bed diameter	6.4 mm (1 ml column) 16 mm (6 ml column)
Bed height	30 mm
Max. pressure	1 ml column: 15 bar (218 psi, 1.5 MPa) 6 ml column: 6 bar (87 psi, 0.6 MPa)
Max. flow rate	1800 cm/h (10 ml/min for 1 ml column) 1800 cm/h (60 ml/min for 6 ml column) in H_20 at 25°C

1 Preparation

Choosing the buffer system

The pH of the start buffer should be at least 0.5 to 1 pH unit above the pI of the target substance when using an anion exchanger (Q) and 0.5 to 1 pH unit below the pI of the target substance when using a cation exchanger (S).

For samples with unknown charge properties, try the following:

- anion exchange (Q)
  start buffer: 20 mM Tris-HCl, pH 8.0
  elution buffer: start buffer including 1 M NaCl, pH 8.0
- cation exchange (S)
  start buffer: 20 mM MES, pH 6.0
  elution buffer: start buffer including 1 M NaCl, pH 6.0

For more information, please refer to the handbook *Ion Exchange Chromatography & Chromatofocusing, Principles and Methods*,

which can be ordered from GE Healthcare or downloaded from our web site.

Preparing buffers and sample

To protect the column and prolong its life, we strongly recommend that you prepare buffers and samples with care. Use water of Milli-Q™ grade or corresponding quality. Degas and filter all buffer solutions through a 0.22 µm filter. Filter the sample. When possible, dissolve or dilute it in start buffer. Buffer exchange and desalting is easily accomplished by gel filtration.

We recommend HiPrep™ 26/10 Desalting, HiTrap™ Desalting or PD-10 Desalting columns.

2 Operation

First time use or after long term storage:

RESOURCE Q is supplied in 20% ethanol and RESOURCE S is supplied in 20% ethanol, 0.2 M sodium acetate.

1. To remove ethanol, wash with 5 column volumes of distilled water at 4 ml/min (RESOURCE 1 ml), 6 ml/min (RESOURCE 6 ml). This step ensures removal of ethanol and avoids the risk of precipitation if buffer salts were to come into contact with the ethanol. The step can be omitted if precipitation is not likely to be a problem.
2. Wash with 5 column volumes of start buffer, at 4 ml/min (RESOURCE 1 ml), 6 ml/min (RESOURCE 6 ml).
3. Wash with 5 column volumes of elution buffer, same flow as step 2.
4. Wash with 5 column volumes of start buffer, same flow as step 2.

Perform a blank run to check conductivity and pH.

Separation by gradient elution

Flow: 4 ml/min (RESOURCE 1 ml), 6 ml/min (RESOURCE 6 ml).

Collect fractions throughout the separations.

1. Equilibrate column with 5-10 column volumes of start buffer or until the baseline, eluent pH and conductivity are stable.
2. Adjust the sample to the chosen starting pH and ionic strength and apply to the column.
3. Wash with 5-10 column volumes of start buffer or until the baseline, eluent pH and conductivity are stable i.e when all unbound material has washed through the column.
4. Begin elution using a gradient volume of 10-20 column volumes and an increasing ionic strength up to 0.5 M NaCl (50%B).
5. Wash with 5 column volumes of 1 M NaCl (100%B) to elute any remaining ionically-bound material.
6. Re-equilibrate with 5-10 column volumes of start buffer or until eluent pH and conductivity reach the required values.

For more information about optimization, please refer to the handbook *Ion Exchange Chromatography & Chromatofocusing, Principles and Methods*.

3 Maintenance

Cleaning

Correct preparation of samples and buffers and application of a high salt wash (1 M NaCl) at the end of each separation should keep most columns in good condition. However, reduced performance, a slow flow rate, increasing back pressure or complete blockage are all indications that the medium needs to be cleaned using more stringent procedures in order to remove contaminants.

The column design does not permit reversing the direction of the flow as it may cause leakage. The numbers of column volumes and time required for each cleaning step may vary according to the degree of contamination.

The following procedure should be satisfactory to remove common contaminants:

1. Wash with at least 2 column volumes of 2 M NaCl at 1 ml/min (RESOURCE 1 ml), 6 ml/min (RESOURCE 6 ml).
2. Wash with at least 4 column volumes of 1 M NaOH (same flow as in step 1).
3. Wash with at least 2 column volumes of 2 M NaCl (same flow as in step 1).
4. Rinse with at least 2 column volumes of distilled water (same flow as in step 1) until the UV-baseline and the eluent pH are stable.
5. Wash with at least 4 column volumes of start buffer or storage buffer (same flow as in step 1) until eluent pH and conductivity have reached the required values.

To remove precipitated proteins, lipids, hydrophobically bound proteins or lipoproteins, refer to the handbook "Ion Exchange Chromatography & Chromatofocusing, Principles and Methods"

Chemical stability

For daily use, RESOURCE columns are stable in all common, aqueous buffers pH 2 to 12, denaturing agents (8 M urea, 6 M guanidine hydrochloride), 75% acetic acid, 1 M NaOH, 1 M HCl, 70% ethanol, 30% acetonitrile and with additives such as nonionic detergents.

Avoid cationic detergents with RESOURCE S. Avoid anionic detergents with RESOURCE Q. Avoid oxidizing agents.

Storage

For column storage, wash with 5 column volumes of distilled water followed by 5 column volumes of 20% ethanol. Include 0.2 M sodium acetate in the 20% ethanol solution for RESOURCE S. Degas the ethanol/water mixture thoroughly and apply at a low flow rate to avoid over-pressuring the column.

Store at 4°C to 30°C. Ensure that the column is sealed well to avoid drying out.

4 Ordering information

Product	Quantity	Code No.
RESOURCE Q, 1 ml	1	17-1177-01
RESOURCE S, 1 ml	1	17-1178-01
RESOURCE Q, 6 ml	1	17-1179-01
RESOURCE S, 6 ml	1	17-1180-01

Accessories	Quantity	Code No.
Union M6 female /1/16" male (for connection to FPLC™ systems)	5	18-3858-01
Fingertight connector1/6", male (for connection to ÄKTA design systems)	10	18-1112-55

Related products	Quantity	Code No.
SOURCE 15Q	10 ml	17-0947-20
SOURCE 15Q	50 ml	17-0947-01
SOURCE 15S	10 ml	17-0944-10
SOURCE 15S	50 ml	17-0944-01
HiTrap Desalting, 5 ml	5 x 5 ml	17-1408-01
HiPrep 26/10 Desalting	1(53 ml)	17-5087-01
HiPrep 26/10 Desalting	4(53 ml)	17-5087-02
PD-10 Desalting columns	30	17-0851-01

Related literature	Code No.
Ion Exchange Chromatography & Chromatofocusing, Principles and Methods	11-0004-21

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