## Anion Exchange columns

TSKgel BioAssist Q
TSKgel DEAE-2SW
TSKgel DEAE-3SW
TSKgel DEAE-5PW
TSKgel DEAE-NPR
TSKgel DNA-NPR
TSKgel DNA-STAT
TSKgel QAE-2SW
TSKgel Q-STAT
TSKgel SAX
TSKgel Sugar AXG
TSKgel Sugar AXI
TSKgel SuperQ-5PW

## Cation Exchange columns

TSKgel BioAssist S
TSKgel CM-2SW
TSKgel CM-3SW
TSKgel CM-5PW
TSKgel CM-STAT
TSKgel OApak-A
TSKgel SCX
TSKgel SP-2SW
TSKgel SP-5PW
TSKgel SP-NPR
TSKgel SP-STAT

## Ion Exchange Tips:

- TSKgel ion exchange columns are offered in glass, PEEK, and stainless steel hardware. Stainless steel (SS) or Pyrex frits are embedded in the body of the column end-fittings of metal and glass columns, respectively. The nominal frit size for SS columns is engraved in the end-fittings; Pyrex frits in the glass columns have a 10 µm nominal pore size.
- Halide salts corrode stainless steel tubing, fitting, and frits. Do not store SS columns in a mobile phase containing NaCl and, where possible, use another salt in the operating buffer. Chlorotrifluorethylene and tetrafluorethylene are the materials in the glass column fittings that come into contact with the mobile phase and sample.
- Good laboratory procedures demand that the analytical column be protected by a guard column. TSKgel guardgel kits, containing column hardware and gel packing, are available to pack your own guard column. In addition, guard cartridges and packed guard columns are available for use with TSKgel ion exchange columns.
- TSKgel ion exchange columns are supplied with an Inspection Data Sheet, which includes a QC chromatogram and test data, and an OCS Sheet summarizing the recommended operating conditions for optimum column performance.
- A separate TSKgel Column Instruction Manual that reviews general guidelines for column installation and care, as well as troubleshooting tips for commonly encountered problems, can be downloaded from the Tosoh Bioscience LLC website (www.tosohbioscience.com).

## About Ion Exchange Chromatography

Ion Exchange Chromatography (IEC) is a technique based on the difference in the strength of the interaction between a sample ion and an oppositely charged functional group on the support. The sample ion competes for the functional group with a counter ion that has been added to the mobile phase as a salt. Elution is most often accomplished by increasing the salt concentration over time.

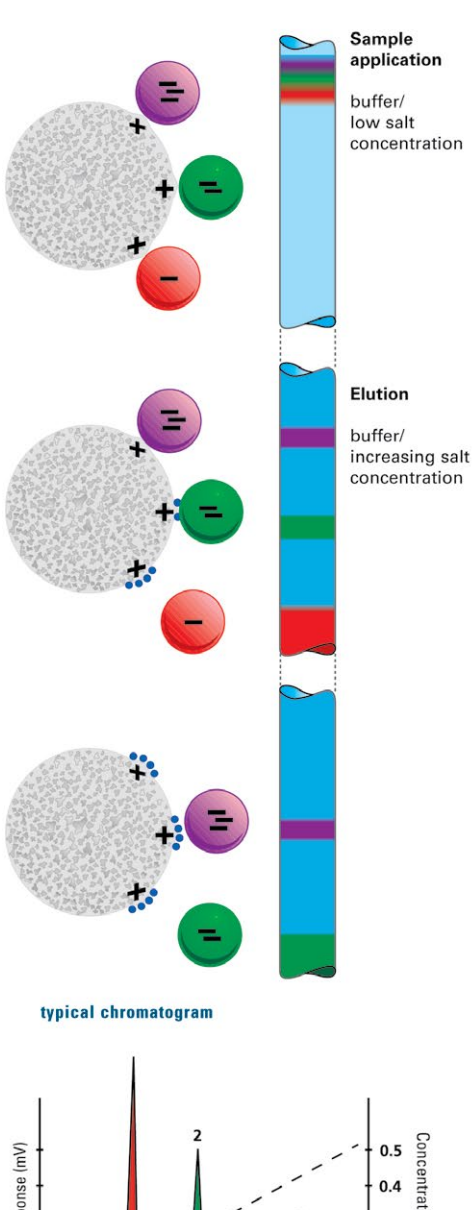
Ion exchange chromatography is the most common separation mode for protein purification schemes. Biomolecules generally have charged groups on their surfaces, which change with the pH of the solution.

Anion Exchange Chromatography is performed with either a strong anion exchange column, containing a quaternary ammonium ion, or with a weak anion exchanger, having either a tertiary or secondary amine functional group, such as DEAE (diethylaminoethyl). A counter ion, often $Cl^-$, maintains electroneutrality.

Cation Exchange Chromatography is performed with either a strong cation exchanger, containing a bonded sulfonic acid group, such as sulfopropyl (SP), or with a weak cation exchanger, containing a weak acid such as carboxymethyl (CM). A counter ion, often $Na^+$, maintains electroneutrality. The advantage of strong vs. weak ion exchangers is that the first are charged over a wider pH range. Weak ion exchangers often provide slightly different selectivity from strong exchangers.

In ion exchange chromatography, the pH of the mobile phase buffer must be between the pI or pKa of the charged molecule and the pKa of the charged groups on the solid support. For example, a molecule with a pI of 8.2 is run in a mobile phase buffer at pH 6.0 with the solid support pKa at 1.2 in cation exchange chromatography. In anion exchange chromatography a molecule with a pI of 6.8 is run in a mobile phase buffer at pH 8.0 with the solid support pKa at 10.3.

*Figure 1: Ion Exchange Chromatography*



## TSKgel Anion and Cation Exchange Chromatography Columns

Tosoh Bioscience offers a broad line of high efficiency columns for analysis and isolation of biomolecules by ion exchange chromatography. Methacrylate, silica, hydrophilic polymer, and polystyrene are used as matrices for the TSKgel line of anion and cation exchange columns. Tables 1 and 2 list the available columns according to matrix and summarize the features and benefits of TSKgel ion exchange columns.

- TSKgel STAT ion exchange columns:
  These are nonporous polymer columns with high surface density of functional groups: quaternary ammonium for anion exchange (Q- and DNA-STAT), carboxymethyl (CM-STAT) and sulfopropyl (SP-STAT) for cation exchange. Particle sizes and dimensions of the TSKgel STAT columns are optimized either for highest throughput or for highest efficiency. Applications for the TSKgel STAT columns include the separation of proteins, protein aggregates, charge isomers of monoclonal antibodies, PEGylated proteins, DNA fragments, nucleic acids, oligo DNA, and siRNA.

- TSKgel DEAE-5PW, SP-5PW, CM-5PW, SuperQ-5PW ion exchange columns:
  The polymethacrylate-based resin, TSKgel 5PW, is a spherical 10 µm particle with approximately 100 nm pores. It is derivatized with diethylaminoethyl (DEAE), sulfopropyl (SP) or carboxymethyl (CM) functionalities to provide a weak anion, a strong cation, and a weak cation exchanger, respectively. The polyamine chemistry employed in TSKgel SuperQ-5PW results in a high capacity strong anion exchanger with a smaller effective pore size than TSKgel DEAE-5PW. Proteins, peptides, DNA- and RNA-derived oligonucleotides, and other nucleic acid fragments are typical samples that are analyzed or isolated on the methacrylate-based TSKgel ion exchange columns.

- TSKgel BioAssist ion exchange columns:
  These columns are also based on methacrylate particle design technology. Particles in TSKgel BioAssist Q columns contain very large pores (~400 nm) that are functionalized with polyamine groups to form a network structure. The capacity of the TSKgel BioAssist Q columns is high over a wide molecular weight range (up to $1.0 \times 10^6$ Da). TSKgel BioAssist S columns are packed with particles possessing 130 nm pores functionalized with sulfopropyl groups. TSKgel BioAssist columns are available exclusively in PEEK housing.

- TSKgel DEAE-NPR, DNA-NPR and SP-NPR ion exchange columns:
  Methacrylate is the backbone of these nonporous resin columns, which are packed with 2.5 µm particles. High column efficiency coupled with low sample capacity restricts the application of these columns to fast analysis and micro-scale preparative isolation. Due to the absence of large pores, protein recovery is generally very high on TSKgel NPR columns.

- TSKgel DEAE-2SW, DEAE-3SW, QAE-2SW, SP-2SW, CM-2SW, CM-3SW ion exchange columns:
  Silica-based TSKgel anion and cation exchange columns with diethylaminoethyl (DEAE), sulfopropyl (SP), trimethylamino (QAE), and carboxymethyl (CM) functional groups are available for analyzing smaller molar mass samples such as nucleotides, drug candidates, catecholamines, and small peptides or proteins. Binding capacity for small to medium size proteins on these columns is approximately double that of the TSKgel 5PW packings due to the smaller pore size and larger surface area.

- Specialty TSKgel polystyrene-based ion exchange columns:
  These columns are available for the analysis of mono- and disaccharides, organic acids and sugar alcohols.

*Table 1: Features and benefits of TSKgel cation exchange columns*

| TSKgel Column Type | Type/Matrix | Benefit |
|---|---|---|
| CM-STAT, SP-STAT | strong(SP-STAT),<br>weak (CM-STAT/polymer | Nonporous with high surface density of carboxymethyl (CM) and sulfopropyl (SP) groups |
| CM-5PW, SP-5PW | strong (SP-5PW),<br>weak (CM-5PW)/polymethacrylate | Polymethacrylate resin derivatized with carboxymethyl (CM) and sulfopropyl (SP) ligands |
| BioAssist S | strong/polymethacrylate | Contain very large pores (130 nm), resulting in high binding capacity and improved recovery of activity; available exclusively in PEEK housing |
| SP-NPR | strong/polymethacrylate | Nonporous with 2.5 µm particles; fast analysis; high protein recovery |
| CM-2SW, CM-3SW, SP-2SW | strong (SP-2SW),<br>weak (CM-2SW, CM-3SW)/silica | Silica-based with carboxymethyl (CM) and sulfopropyl (SP) functional groups |
| SCX, OApak-A | strong (SCX),<br>weak (OApak-A)/ polymethacrylate | Specialty columns for the analysis of organic acids, saccharides and alcohols |

*Table 2: Features and benefits of TSKgel anion exchange columns*

| TSKgel Column Type | Type/Matrix | Benefit |
|---|---|---|
| Q-STAT, DNA-STAT | strong (Q-STAT),<br>weak (DNA-STAT)/polymer | Nonporous with high surface density of quaternary ammonium groups |
| DEAE-5PW, SuperQ-5PW | strong (SuperQ-5PW),<br>weak (DEAE-5PW)/polymethacrylate | Polymethacrylate resin derivatized with diethylaminoethyl (DEAE) and trimethylamino (SuperQ) ligands |
| BioAssist Q | strong/polymethacrylate | Contain very large pores (400 nm), resulting in high binding capacity and improved recovery of activity; available exclusively in PEEK housing |
| DEAE-NPR, DNA-NPR | weak/polymethacrylate | Nonporous with 2.5 µm particles; fast analysis; high protein recovery |
| DEAE-2SW, DEAE-3SW, QAE-2SW | strong ( QAE-2SW),<br>weak (DEAE-2SW, DEAE-3SW)/silica | Silica-based with diethylaminoethyl (DEAE), and trimethylamino (QAE) functional groups |
| Sugar AXG, Suger AXI, SAX | strong/polystyrene | Specialty columns for the analysis of mono and disaccharides, as well as organic acids and sugar alcohols |