TSKgel® SEC Columns for Biomolecules

Modern molecular exclusion solutions for (U)HPLC

TSKgel® SuperSW series
TSKgel® UltraSW Aggregate
TSKgel® UP-SW series

High-performance, high-resolution columns
2 to 4 micron particles increase resolution and sensitivity
Low non-specific adsorption
High reproducibility due to a well-defined pore size
Complete range of columns for protein analysis,
mABs, fragments and agglomerates
HPLC and UHPLC applications

TOSOH BIOSCIENCE

## What kinds of biomolecules are we interested in?

- Recombinant therapeutic proteins
- Monoclonal antibodies (mAbs)
- Antibody derivatives

The appearance of by-products in the synthesis of these biomolecules generates a loss of therapeutic efficacy of these species and immunogenesis. Therefore, they must be analyzed and, in their presence, purified.
The by-products to be analyzed are mAbs fragments and protein agglomerates.

Complete Protein   Fragments

agglomerates
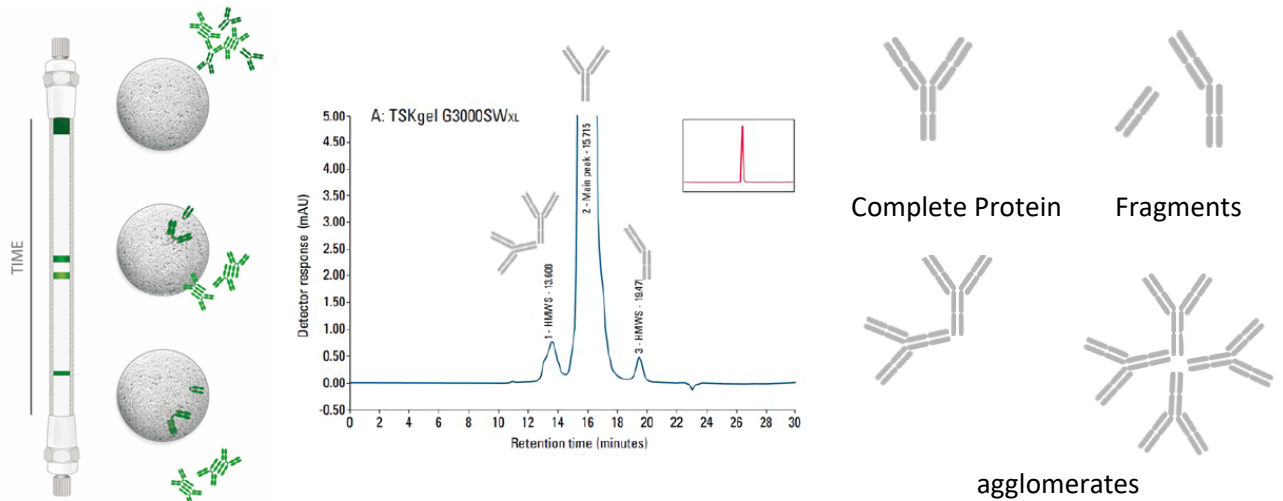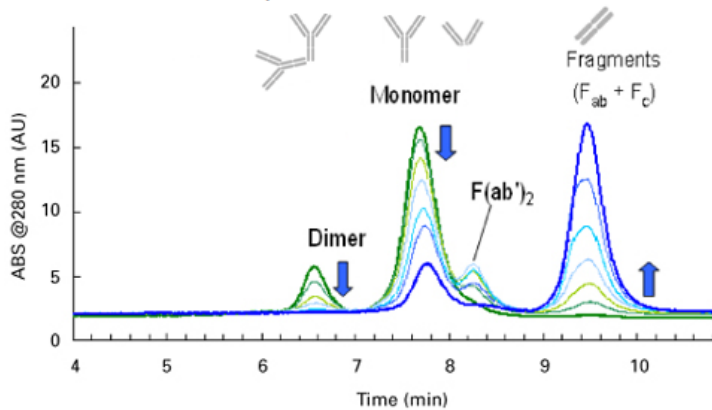
Size separation of IgG components from Papain digestion

Range of available columns

	Material base:	Particle Size (average):	Pore Size (average):	Functional Group:	pH Stability:	Calibration Range (globular proteins):
TSK gel Super SW2000	Silica	4 µm	12,5 nm	Diol	2.5 - 7.5	5,000 - 150,000 Da
TSK gel Super SW3000	Silica	4 µm	25,0 nm	Diol	2.5 - 7.5	10,000 - 500,000 Da
TSKgel SuperSW mAb HR	Silica	4 µm	25,0 nm	Diol	2.5 - 7.5	10,000 - 500,000 Da
TSK gel SuperSW mAb HTP	Silica	4 µm	25,0 nm	Diol	2.5 - 7.5	10,000 - 500,000 Da
TSKgel UltraSW Aggregate	Silica	3 µm	30,0 nm	Diol	2.5 - 7.5	10,000 - 2,000,000 Da
TSKgel UP-SW2000	silica	2 µm	12,5 nm	Diol	2.5 - 7.5	1,000 - 150,000 Da
TSKgel UP-SW3000	Silica	2 µm	25,0 nm	Diol	2.5 - 7.5	10,000-500,000 Da

Range of available columns

Separation of mAbs fragments
HPLC
SuperSW 2000   4 μm 12,5 nm
UHPLC
UP-SW2000   2 μm 12,5 nm

Separation of mAbs and proteins (up to 200K Da)
HPLC
SuperSW mAb HR   4 μm 25.0 nm 30cm*7.8mm
SuperSW mAb HTP   4 μm 25.0 nm 15cm*4.6mm
UHPLC
UP-SW3000   2 μm 25.0 nm
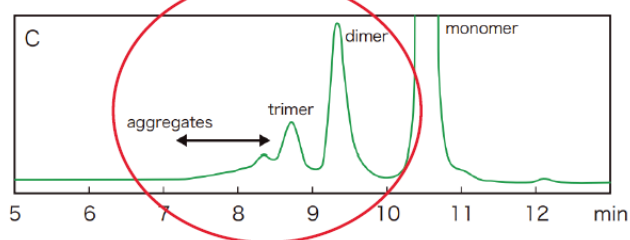
Separation of mAbs aggregates
HPLC
Ultra SW Aggregate   3 μm 30.0 nm 30cm*7.8mm
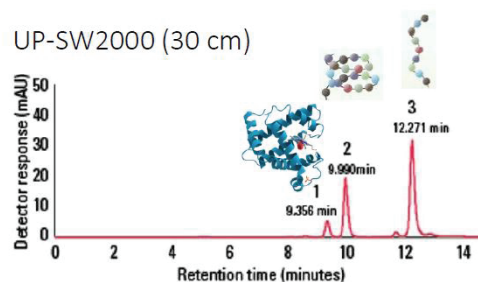**UHPLC**
UP-SW Aggregate   3 μm 30.0 nm 30cm*4.6mm

Analysis of Erbitux aggregates, chimeric mAB with 30 nm particles by UHPLC

Samples: 1: Myoglobin 16.7 kDa, 2: 58 aa, 6.5 kDa, 3: 14 aa, 1.6 kDa

Separation of myoglobin and other smaller peptides by UHPLC

Main benefits:
Increased resolution and throughput of well known the G2000SWxl and G3000SWxl
Columns with excellent robustness and batch-to-batch reproducibility
Resolutions of more than 30,000 plates assured (30cm)
Wide range of columns for all requirements
Technical support from expert biochromatographers for method optimization

Technical Note

UHPLCs are already designed with very small dead volumes to ensure good efficiency, but in HPLCs, if we want to get good results with these high resolution columns, we must optimize our chromatographic system:

CONNECTIONS
Conventional 0.1 mm tubing can be used, but the length should be as short as possible.
The dead volume between the column and the detector cell should be less than 20 μL.

INJECTOR
The best results are obtained with a low-diffusion manual injector (Rheodyne 8152).
The output dead volume of the autosampler should be as low as possible.

SAMPLE VOLUME
The sample volume should be 10 μl or less. The sample loading should be less than 100 μg
(for a 4,6 mm column bore).

PRECOLUMN
A precolumn or in-line filter is strongly recommended to reduce clogging and contamination.

DETECTOR
For best results, use a flow cell with a maximum of 2 μL.
The 2 μL flow cell will provide the highest efficiencies.
A 2-10 μL flow cell can be used for columns of 4.6 mm inner diameter.
However, the theoretical plates will be reduced.
A small acquisition time constant (less than 0.5 seconds) is needed to achieve the best column performance.

PUMP
A pump with an accurate delivery rate between 0.01 mL/min and 0.35 mL/min is recommended.

Which column is the right one?

Instrument available or updated?

Molecular size?

Node	Connection
UP-SW2000	UHPLC
SuperSW 2000	HPLC
UP-SW3000	UHPLC
SuperSW mAb	HPLC
UP-SW Aggregate	UHPLC
Ultra SW Aggregate	HPLC

Pore size of 12.5 nm is perfect

I need to improve the resolution over...

Pore size of 25 nm is perfect

I need to improve the resolution over...

Pore size of 30 nm is perfect

I need to improve the resolution over...

Tosoh Bioscience provides column for the analysis of a diversity of molecules: Oligos, mRNA, recombinant proteins, vitamins.

Contact Teknokroma for more information

Which proteins are analysed?

mAb or 10-100 kDa fragments

mAb or 50-200 kDa

mAb aggregates or 100-1000 kDa

Non-proteins