

# TSKgel® SuperSW mAb HTP/HR TSKgel® UltraSW AGGREGATE

#442

## INTRODUCTION

Aqueous size exclusion chromatography (SEC) is the method of choice for the analysis of protein fragments, monomers, and aggregates under non-denaturing conditions. Based on the flow of the sample through a porous stationary phase SEC separates molecules according to their size, or more precisely, their hydrodynamic volume. In aqueous elution systems SEC is also referred to as gel filtration chromatography (GFC). TSKgel G3000SW<sub>XL</sub> columns have been the industry's standard for quality control of monoclonals by SEC for decades. Based on the proven proprietary surface technology of the renowned TSKgel SW series, a new series of silica-based SEC columns was engineered to provide shorter analysis time or higher resolution for antibody analysis.

## HIGHLIGHTS

- Optimized for antibody analysis
- Small particle size for UHPLC use
- Highest resolution with SuperSW mAb HR
- Fast separation with SuperSW mAb HTP
- Higher molecular weight range with UltraSW Aggregate

## FEATURES

The new series of dedicated SEC columns for mAb analysis delivers significant advancements in resolution. It comprises of three different columns and their guards. Table 1 summarizes the characteristics of the mAb SEC columns and Figure 1 shows the calibration curves and the molecular weight range of the three columns.

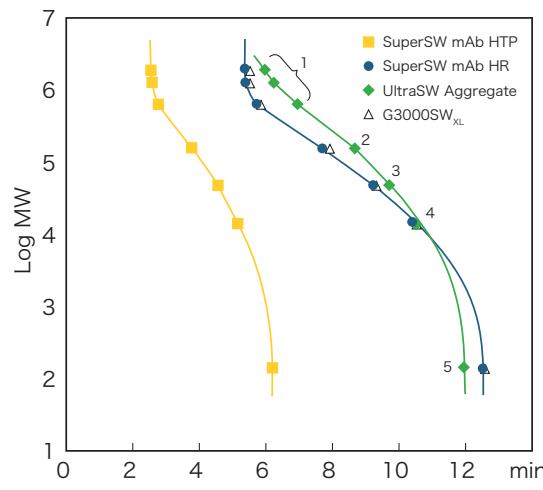
## CHARACTERISTICS OF TSKgel mAb COLUMNS

Column	TSKgel SuperSW mAb HR	TSKgel SuperSW mAb HTP	TSKgel UltraSW Aggregate
Dimension	7.8 mm ID x 30 cm	4.6 mm ID x 15 cm	7.8 mm ID x 30 cm
Theoretical plates	≥30.000	≥15.000	≥35.000
Base material	Silica gel		Silica gel
Particle size	4 µm		3 µm
Separation range (globular proteins)	10,000 - 500,000 Da		10,000 - 2,000,000 Da

➤ Table 1

The calibration curve of TSKgel SuperSW mAb HR is most similar to the one of TSKgel G3000SW<sub>XL</sub>, the current industrial standard for antibody analysis. The new columns are highly reproducible. The high Batch-to-Batch stability is proved in Figure 2 for TSKgel Super SW mAb HR. The lifetime of the columns can be further improved when using the corresponding guard columns.

## CALIBRATION CURVE



➤ Figure 1

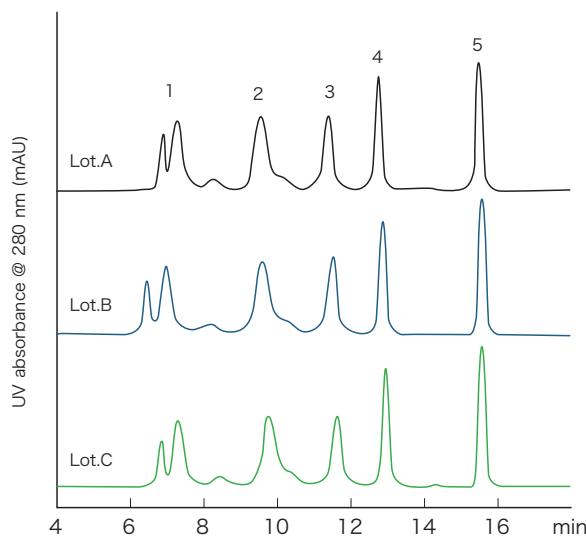
Columns: TSKgel SuperSW mAb HTP 4.6 mm ID x 15 cm, TSKgel SuperSW mAb HR, TSKgel UltraSW Aggregate, TSKgel G3000SW<sub>XL</sub> (all 7.8 mm ID x 30 cm)  
Mobile phase: 0.2 mol/L phosphate buffer, pH 6.7 0.05% Na<sub>3</sub>O; Flow rate: 1.0 mL/min, 0.35 mL/min (SuperSWmAbHTP); Temperature: 25°C; Detection: UV 280 nm; Inj. vol.: 10 µL, 5 µL (SuperSW mAb HTP )  
1. Thyroglobulin (MW 640,000), 2. γ-Globulin (MW 155,000), 3. Ovalbumin (MW 47,000), 4. Ribonuclease A (MW 13,700)  
5. p-Aminobenzoic acid (MW 137)



Each of the new SEC columns is tailored to a specific separation problem. TSKgel SuperSW mAb HTP - "HTP" standing for high throughput - was developed to enable an easy transfer of HPLC methods based on TSKgel SW<sub>XL</sub> to fast UHPLC analysis. Small particle size silica beads are packed in UHPLC column hardware with 4.6 mm inner diameter. This enables to double the throughput without compromising resolution (Figure 3).

TSKgel SuperSW mAb HR - "HR" indicating high resolution - delivers superior resolution over the whole range from fragments to aggregates when analysing monoclonals (Figure 4). It has the same column dimensions - 7.8 mm by 30 cm length - as the established TSKgel G3000SW<sub>XL</sub>. TSKgel Ultra SW Aggregate (7.8 mm ID x 30 cm) features a smaller particle size and a higher exclusion limit. It offers a wider separation window in the aggregate region (Figure 4C).

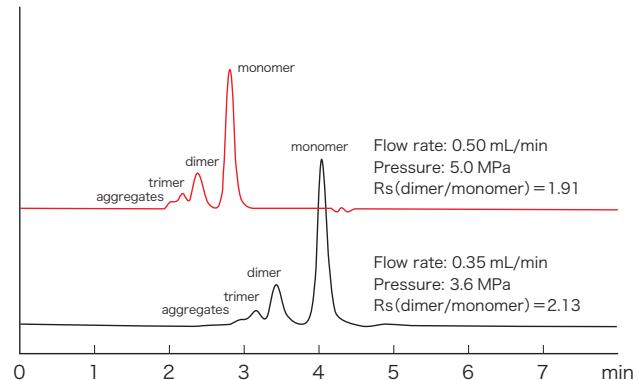
#### BATCH TO BATCH REPRODUCIBILITY



**FIGURE 2**

Column: TSKgel SuperSW mAb HR (7.8 mm ID x 30 cm)  
Eluent: 0.2 mol/L phosphate buffer (pH 6.7)+ 0.05%  $\text{NaN}_3$   
Flow rate: 0.8 mL/min; Detection: UV @ 280 nm; Inj. volume: 10  $\mu\text{L}$ ; Sample: 1. Thyrobulobulin, 2.  $\gamma$ -Globulin, 3. Ovalbumin, 4. Ribonuclease A, 5. p-Aminobenzoic acid

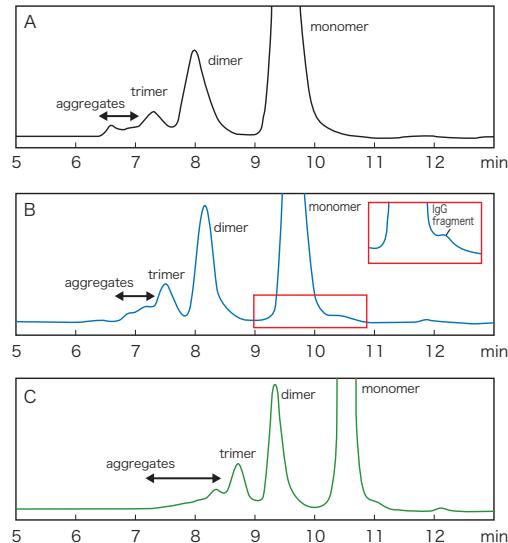
#### FAST ANALYSIS OF mAb AGGREGATION



**FIGURE 3**

Column: TSKgel SuperSW mAb HTP (4.6 mm ID x 15 cm)  
Elution: 0.2 mol/L phosphate buffer (pH 6.7) + 0.05%  $\text{NaN}_3$   
Flow rate: 0.50 mL/min, 0.35 mL/min; Detection: UV @ 280 nm  
Temp.: 25°C; Sample: monoclonal antibody (mouse-human chimeric IgG, Erbitux), 5  $\mu\text{L}$

#### COMPARISON OF AGGREGATE ANALYSIS



**FIGURE 4**

Columns: A. TSKgel G3000SW<sub>XL</sub>, B. TSKgel SuperSW mAb HR, C. TSKgel UltraSW Aggregate; Dimension: 7.8 mm ID x 30 cm; Eluent: 0.2 mol/L phosphate buffer (pH 6.7) + 0.05%  $\text{NaN}_3$ ; Flow rate: 0.8 mL/min; Detection: UV @ 280 nm; Temp.: 25°C; Sample: monoclonal antibody, (mouse-human chimeric IgG, Erbitux), 10  $\mu\text{L}$

## Ordering information

Part-No	Description	Matrix	Housing	Dimensions
22854	TSKgel SuperSW mAb HR, 4 µm, 250 Å	Silica	Stainless steel	7.8 mm ID x 30.0 cm L
22855	TSKgel SuperSW mAb HTP, 4 µm, 250 Å	Silica	Stainless steel	4.6 mm ID x 15.0 cm L
22856	TSKgel UltraSW Aggregate, 3 µm, 300 Å	Silica	Stainless steel	7.8 mm ID x 30.0 cm L
22857	TSKgel Guardcolumn SuperSW mAb, 4 µm	Silica	Stainless steel	6.0 mm ID x 4.0 cm L
22858	TSKgel Guardcolumn SuperSW mAb, 4 µm	Silica	Stainless steel	3.0 mm ID x 2.0 cm L
22859	TSKgel Guardcolumn UltraSW, 3 µm	Silica	Stainless steel	6.0 mm ID x 4.0 cm L

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