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SEC (Gel filtration) HPLC Low Molecule Weight (LMW) Protein Standard

Each tube contains protein mixture lyophilized from 100 μ L of standard solution

Total 2 tubes

Product Number: **CM92005**

Product Description

CellMosaic's size exclusion chromatography (SEC) HPLC LMW protein standard is designed for day to day use to check HPLC column performance and the analysis of bioconjugates. The kit consists of four proteins with a MW range of 6.5 to 43 KDa and a small molecule with MW of 154 Da. The broad range of protein molecular weight coverage allows easy identification of small molecules, conjugates, and aggregates in a bioconjugation mixture, such as protein and peptide labeling and conjugation. CellMosaic routinely uses this product for its internal bioconjugation-related research.

A total of two micro-centrifuge tubes are in a package. Each micro-centrifuge tube contains protein mixture lyophilized from 100 μ L of standard solution. The product is ready for use after reconstituting in 100 μ L of deionized water.

Application of the Product

- Calibration of gel filtration and size exclusion column
- Estimate the size and molecular mass of proteins
- Analysis of small molecules, bioconjugates, and aggregates

Key Features of the Product

- Lyophilized solid and ready to use for HPLC analysis after dissolving in water
- MW coverage: 154 Da to 43 KDa

Composition

#	Name	MW (Da)
1	2,5-dihydroxybenzoic acid	154
2	Aprotinin	6,511
3	RNase A	13,700
4	Carbonic anhydrase	29,000
5	Ovalbumin (albumin chicken egg)	43,000

Storage/Stability

- Once reconstitute, CellMosaic's protein standard solution is stable at RT for months.
Recommended storage of the product is below -20°C, which is viable for several years without any sign of decomposition.

Procedure

1. Take one tube out and warm to ambient temperature.
2. Add 100 μ L of deionized water and vortex for 30 seconds to dissolve.
3. Centrifuge briefly to ensure no liquid is in the cap. Transfer the supernatant to an HPLC vial.
4. Inject 8 μ L for HPLC analysis (Note: the amount is optimized for a standard SEC HPLC analytical column with a volume of 10-15 mL. For lower or higher column volumes, scale the injection volume accordingly.)

Application Note

Calibration of Protein and Conjugate MW

The approximate MW of an unknown protein or protein conjugate can be determined by comparing its retention time to the protein standard. **Figure 1** gives an example of the SEC profile. For more accurate calculation, an SEC MW standard curve is recommended (**Figure 2**).

Figure 1. SEC HPLC profile of protein standards in a TSKgel BioAssist G3000SW_{XL} (7.8 mm x 30 cm, 5 μ m, Part No. 08541). HPLC condition: isocratic PBS buffer at 0.5 mL/min. Instrument: Agilent 1100 HPLC series.

#	Name	MW (KDa)	Rt (min)
1	2,5-dihydroxybenzoic acid	154	26.364
2	Aprotinin	6,511	23.708
3	RNase A	13,700	21.592
4	Carbonic anhydrase	29,000	20.055
5	Ovalbumin (albumin chicken egg)	43,000	17.978

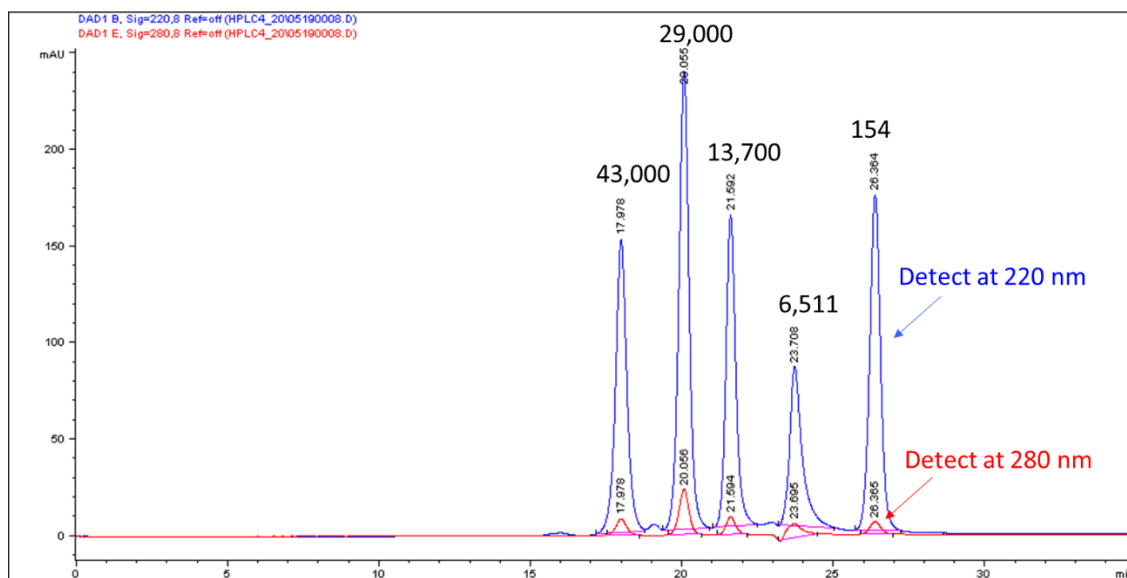
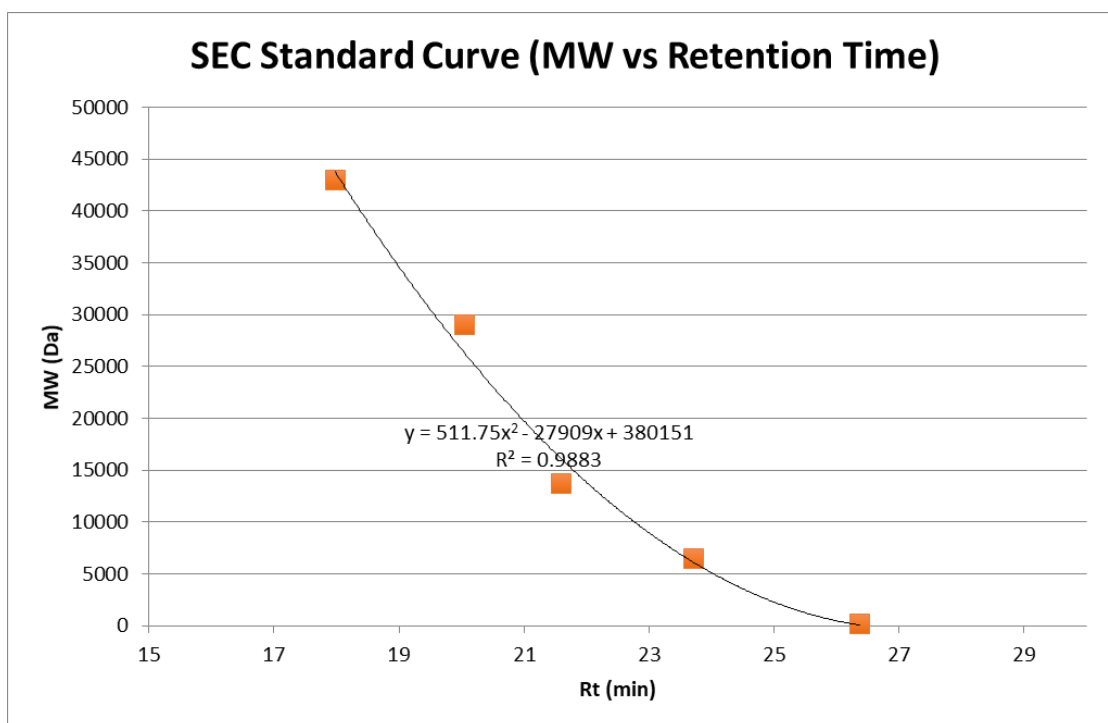


Figure 2. SEC MW standard curve of TSKgel BioAssist G3SW_{xl} (7.8 mm x 300 mm, Part No. 0008541). Data from Figure 1.



Assessing Column Performance

The protein standard mixture can be used to assess the column performance. The SEC profile of the protein standards may differ from column to column after some use. The quality of the SEC profile will indicate whether the column is good. For example, split peaks or tailing may indicate column resin-bound hydrophobic small molecules and that the column needs cleaning.

Important Notes & Contact Information

READ BEFORE USING ANY RESOURCES PROVIDED HEREIN

The information provided in this document and the methods included in this package are for information purposes only. CellMosaic provides no warranty of performance or suitability for the purpose described herein.

Sample data are provided for illustrative and example purposes only and represent a small dataset used to verify kit performance in the CellMosaic laboratory. Information about the chemicals and reagents used in the kit are provided as necessary.

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