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APPLICATION NOTE

Determining Lentinan Molecular Weight Using the BeSEC

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Introduction

Lentinan is a bioactive polysaccharide extracted from shiitake mushrooms, widely recognized for its immunomodulatory effects. It is commonly used as an adjuvant in cancer therapy to ease the side effects of chemotherapy and radiotherapy. Beyond oncology, lentinan exhibits antiviral and antioxidant activity and is frequently incorporated into health supplements to enhance immunity and slow aging. In food applications, it improves texture and stability while adding nutritional value.

Molecular weight plays a critical role in lentinan's functionality. Higher molecular weight molecules tend to adopt more ordered structures and bind more effectively to immune cell receptors, resulting in stronger immunomodulatory effects. Lower molecular weight molecules dissolve and absorb more easily, but their biological activity may be shorter-lived.

Selecting the appropriate molecular weight is essential for optimizing performance in specific applications.

Advanced Light Scattering Detector BeSEC

Experimental Section

This study utilized a Size Exclusion Chromatography (SEC) system equipped with refractive index (RI) and light scattering (LS) detectors. The light scattering detector is the BeSEC LS2 from Bettersize Instruments, with 90° and 7° angles. The BeSEC workstation combines light scattering with RI or UV signals to calculate molecular weight averages (Mn, Mw and Mz) and distributions.

System Configuration:

- Detectors: Light Scattering (LS) + Refractive Index (RI)
- Column: Shodex GPC KF-806M
- Mobile phase: 0.05 M $NaNO_3$ aqueous solution
- Flow rate: 0.7 mL/min
- Injection volume: 100 µL
- Column temperature: 40 °C
- dn/dc: 0.129 mL/g

Sample Preparation:

Five lentinan samples were analyzed. Each powder was accurately weighed and dispersed in 0.05 M $NaNO_3$, stirred until clear (1 to 3 mg/mL), filtered through a 0.22 µm PES syringe filter, and transferred into vials and placed in the autosampler for measurement.

Results and Discussion

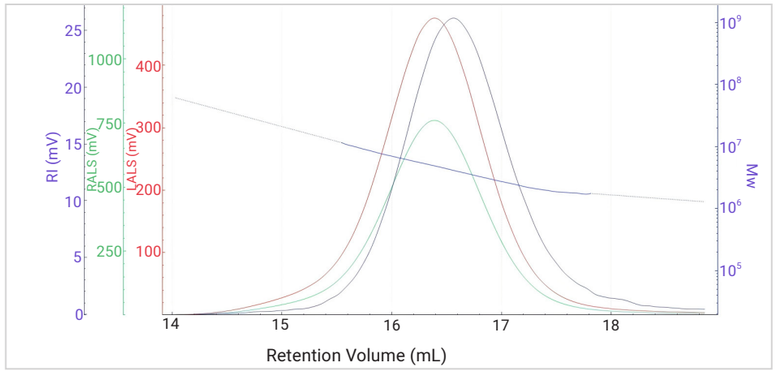

Figure 1. Elution profiles of the multi-detector signals (top) and molecular weight distribution (bottom) for Sample A

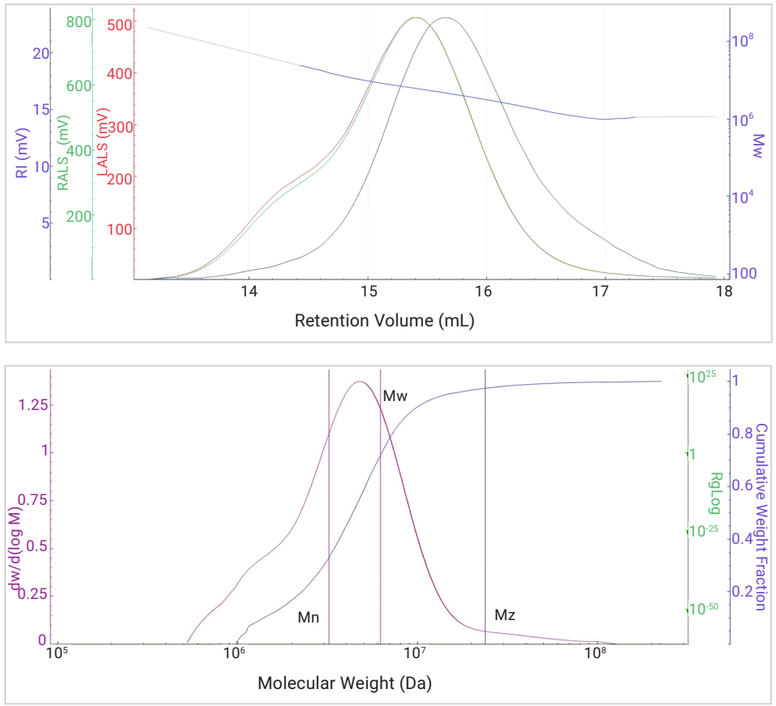

Figure 2. Elution profiles of the multi-detector signals (top) and molecular weight distribution (bottom) for Sample B

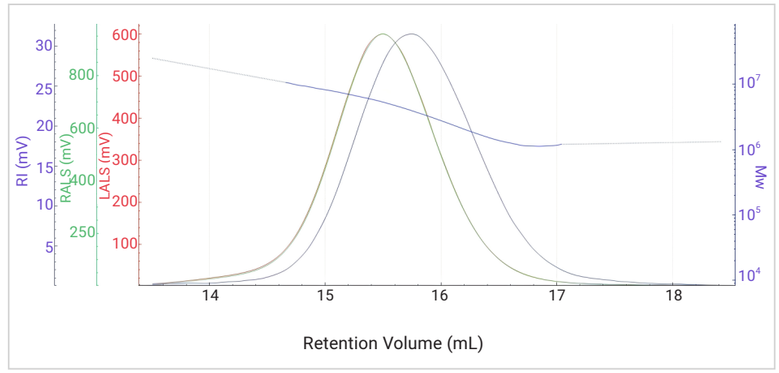

Figure 3. Elution profiles of the multi-detector signals (top) and molecular weight distribution (bottom) for Sample C

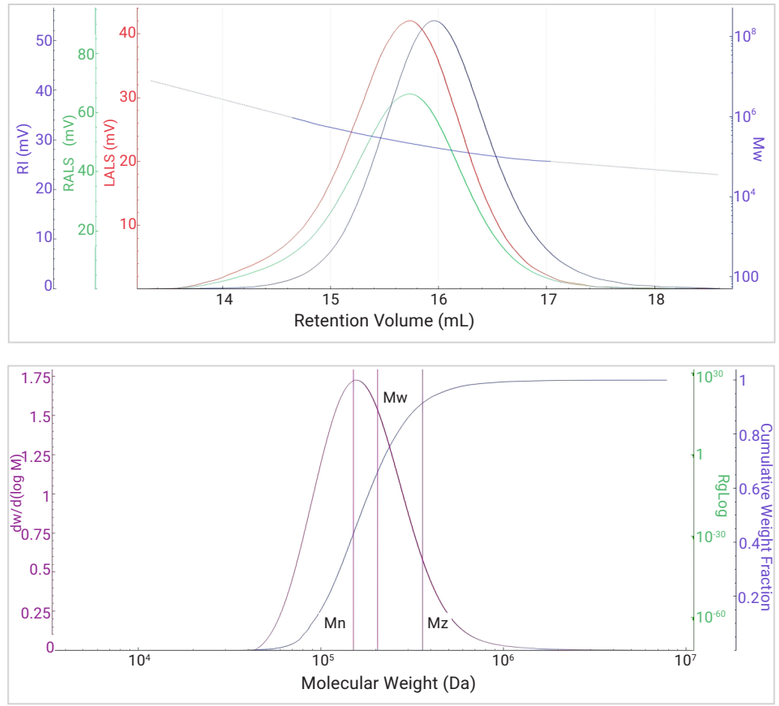

Figure 4. Elution profiles of the multi-detector signals (top) and molecular weight distribution (bottom) for Sample D

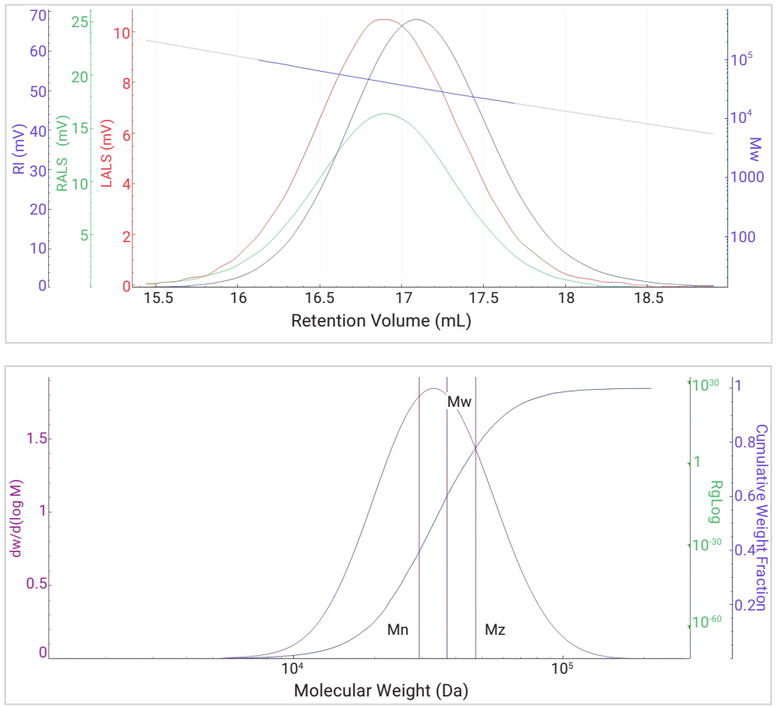

Figure 5. Elution profiles of the multi-detector signals (top) and molecular weight distribution (bottom) for Sample E

Figures 1 to 5 show chromatograms of the five lentinan samples. The refractive index (RI) signal is shown in blue, the right-angle light scattering (RALS) signal in green, and the low-angle light scattering (LALS) signal in red. The molecular weight distribution, plotted as a purple line, represents molecular weight as a function of elution time.

Across all samples, the signals are clean with flat baselines, minimal noise, and strong signal-to-noise ratios. In Figure 1, the molecular weight profile decreases steadily with elution volume, consistent with SEC principles, larger species eluting first followed by smaller ones. Scattering peaks exhibit no tailing and the molecular weight curve remains stable at the end, indicating effective size-based separation.

Table 1. Molecular weight results of lentinan samples

No.	Mn (kDa)	Mw (kDa)	Mz (kDa)	Mw/Mn
Sample A	3559	4465	6128	1.25
Sample B	3218	6222	23741	1.37
Sample C	2903	4168	5676	1.13
Sample D	150.8	204.5	360.4	1.35
Sample E	29.2	37.1	47.5	1.27

Table 1 summarizes the molecular weight results for all five samples. Significant differences in molecular weight were observed, which directly impact solubility, viscosity, biological activity, and formulation behavior. Consequently, each sample may be better suited for different applications.

Conclusion

This study demonstrates the use of the BeSEC LS2 with light scattering detection to characterize the molecular weight of lentinan samples. The results reveal substantial variation in molecular weight among the five samples, providing critical insight for selecting the appropriate grade for pharmaceutical, nutraceutical, or food applications.

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