Uniformity & Reproducibility in Response to Polysaccharides

Varian ELSD

Advantage Statement: The Varian Evaporative Light Scattering Detector provides highly consistent results from injection to injection in peak area response, which is independent of the molecular weight of the sample.

When quantifying a sample using a calibration of response versus concentration, it is important to be able to generate accurate and reproducible peak areas. To illustrate the peak area reproducibility of the Varian ELSD, a test was designed based on the run-to-run reproducibility of a range of narrow polysaccharide standards of different molecular weight values, chosen to illustrate the performance of the detector as a function of molecular weight. Direct injections were made to eliminate variability associated with column efficiency or other chromatographic parameters. The system was configured with a 1 m length of 0.01 in. internal diameter stainless steel tubing directly connecting the injection valve and the detector to give a slight delay between injection and detection. A range of pullulan polysaccharide standards, also from Varian, Inc., was prepared at 0.2 mg/mL and allowed to dissolve for at least two hours. Each sample was injected 12 times.

Samples: Pullulan polysaccharides
(Mp 11800, 47300, 404000 and 788000)
Sample Concentration: 0.2 mg/mL
Column: Direct injection
Eluent: Water
Flow Rate: 1.0 mL/min
Injection Vol: 50 µL
Detection: Varian ELSD (evap=120 °C, neb=90 °C, gas=1.5 SLM)

Figure 1 is an example chromatogram and Table 1 shows the peak areas and the percentage variation in peak area obtained for 12 consecutive repeat injections for four different molecular weight polysaccharides.

The results show that the percentage variation is of the order of 1 %, indicating that the response of the Varian ELSD is consistent from injection to injection in peak area response and is independent of the molecular weight of the sample.

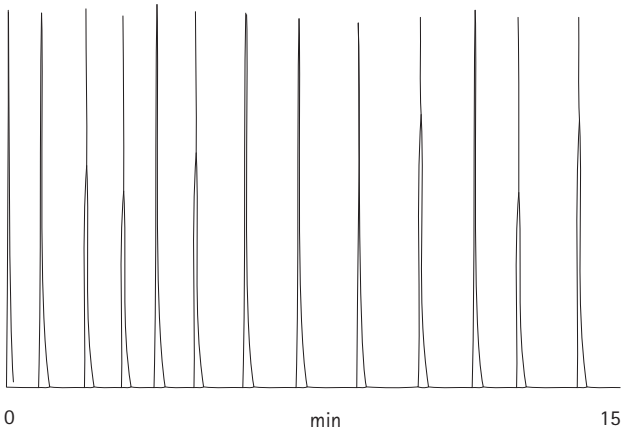

Figure 1. Example of the repeat injections of Mp 788,000.

Table 1. % variation in peak areas for 12 direct injections of polysaccharide samples.

Peak Areas / mV.s				
Injection No.	Mp 11800	Mp 47300	Mp 404000	Mp 788000
1	344.40	346.58	336.8309	371.9879
2	340.00	344.00	340.7170	372.4528
3	344.43	347.48	336.1602	368.4118
4	345.17	348.13	339.3797	370.2734
5	339.90	350.99	337.2607	366.9918
6	345.75	347.38	344.5639	373.4639
7	341.93	348.28	340.9691	367.9548
8	341.35	348.95	347.3055	366.6957
9	344.54	350.5	343.4333	367.3282
10	347.18	353.86	338.6402	367.8734
11	344.44	349.58	344.4332	366.252
12	348.87	358.98	345.5987	366.252
Mean	344.0	349.6	341.3	368.9
Std Dev.	2.8	3.8	3.7	2.5
% Variation	0.8	1.0	1.1	0.7

These data represent typical results. For further information, contact your local Varian Sales office.

Varian, Inc.
www.varianinc.com
North America: 800.926.3000, 925.939.2400
Europe The Netherlands: 31.118.67.1000
Asia Pacific Australia: 613.9560.7133
Latin America Brazil: 55.11.3238.0400
Other sales offices and dealers throughout the world—check our Web site.

TB1034 SI-1726/A/9.08 Printed in the UK