

Method Development for ADC Kadcyra by Size Exclusion LC Using a Biozen™ dSEC-2 Column

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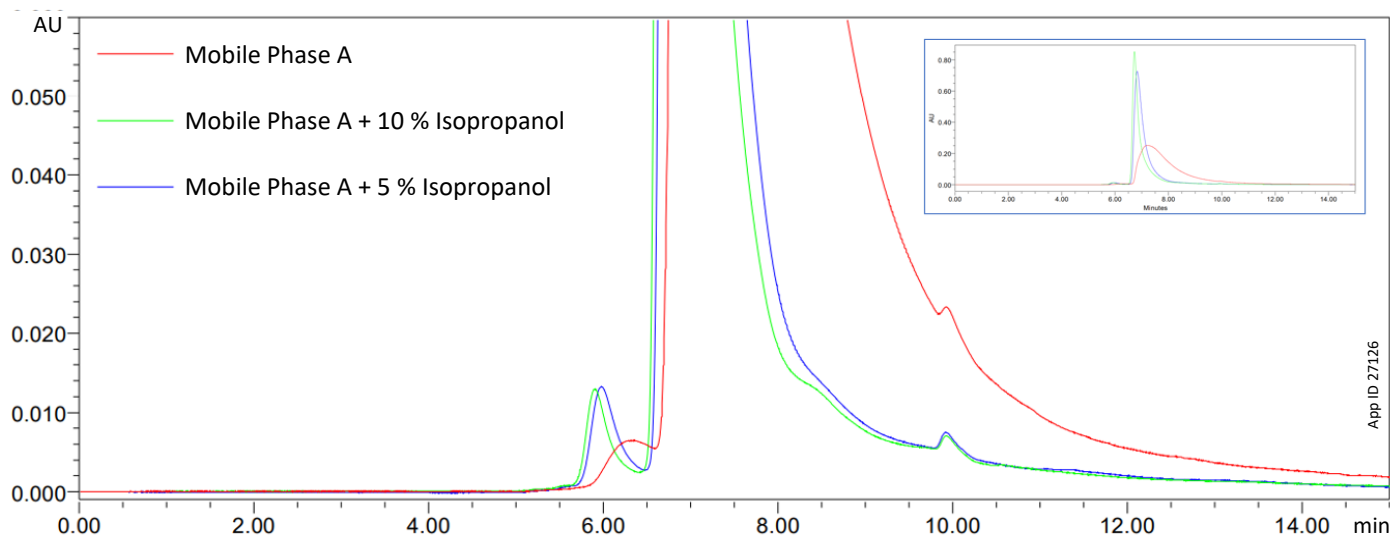
Overview

Size Exclusion Chromatography (SEC) is a technique for the separation of biomolecules based on their size in solution is particularly useful for quantitating size variants and high molecular weight aggregates. This is a common analytical method for monoclonal antibodies (mAbs), as aggregate can be quantitated by percentage of peak area. In this application note, we explore method development using standard phosphate containing buffers with different percentages of Isopropanol for aggregate analysis of an antibody-drug conjugate (ADC), Kadcyra. The use of 10 % Isopropanol was necessary in the efficiency of separating aggregate from the ADC.

LC-UV Conditions

Column: Biozen 1.8 μ m dSEC-2
Dimension: 300 x 4.6 mm
Part No.: [00H-4787-E0](#)
Mobile Phase: A: 200 mM Potassium Phosphate + 250 mM Potassium Chloride, pH 6.2
B: Mobile Phase A + 5 % Isopropanol
C: Mobile Phase A + 10 % Isopropanol
Flow Rate: 0.35 mL/min
Injection Volume: 1 μ L
Temperature: 25 $^{\circ}$ C
Detection: UV @ 214 nm
Sample: Kadcyra (10 μ g)

Figure 1. Comparison of the ADC Kadcyra, with Three Different Mobile Phases Using the Biozen 1.8 μ m dSEC-2 Column. The Addition of 10 % Isopropanol (Green) successfully separated the aggregate from the ADC.



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