

# Improved Sensitivity and Recovery Using the Biozen™ dSEC-2 Column

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### **Overview**

Size exclusion is an important technique in the quantitation of aggregate formation for monoclonal antibodies, a critical quality attribute required for therapeutic antibodies. The Biozen dSEC-2 column shows improved recovery and greater sensitivity over more traditional column technologies attributed to its hydrophilic inert surface chemistry. In this application note, we show that peak area was higher for the Biozen dSEC-2 column showing less adsorption is occurring. In addition, we demonstrate superior peak shape and recovery under USP mobile phase conditions with the Biozen dSEC-2 column.

# **LC Conditions**

Columns: Biozen 1.8 μm dSEC-2 (00H-4787-E0)

Waters® XBridge® 2.5 μm Premier Protein SEC Tosoh Bioscience® TSKgel® 2.0 μm UP-SW3000

Dimensions: 300 x 4.6 mm (Biozen)

300 x 4.6 mm (Waters) 300 x 4.6 mm (Tosoh)

Mobile Phase: 200 mM Potassium Phosphate + 250 mM Potassium

Chloride, pH 6.2

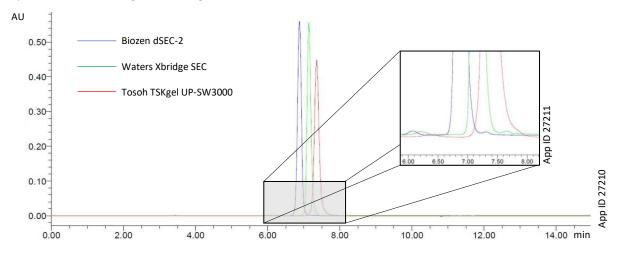
Flow Rate: 0.35 mL/min

Injection Volume:  $3 \mu L$ Temperature:  $25 \,^{\circ}C$ 

Instrument: Waters ACQUITY® UPLC H-Class

Detection: UV @ 280 nm

Figure 1. Representative Chromatogram of 0.5 mg/mL of Trastuzumab on Three Different Columns.



Name		Retention Time (min)	% Area	Area	Height
Biozen dSEC-2	Aggregate	6.07	0.25	10960	1019
	Trastuzumab	6.88	99.61	4320932	560220
	Fragment 1	7.30	0.14	6045	713
Waters XBridge SEC	Aggregate	6.21	0.19	8122	750
	Trastuzumab	7.14	99.7	4268340	566107
	Fragment 1	7.65	0.11	4748	660
Tosoh TSKgel UP-SW3000	Aggregate	6.38	0.2	8513	617
	Trastuzumab	7.36	99.8	4221284	448009

Fragment 1

Not Seen

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