Analysis of Intact Proteins on a Thermo Scientific Accucore 150-C4 150 Å Pore Diameter NanoLC Column

Application #379

24 August 2012

Analysis of Intact Proteins on a Thermo Scientific Accucore 150-C4 150 Å Pore Diameter NanoLC Column

General Information

Market: Biopharmaceutical
Matrix: Standard
Instrument type: UHPLC

Description

Accucore HPLC columns use Core Enhanced Technology to facilitate fast and high efficiency separations. The 2.6 µm diameter particles have a solid core and a porous outer layer. The optimized phase bonding creates a series of high coverage, robust phases. The tightly controlled 2.6µm diameter of accucore particles results in much lower backpressures than typically seen with 2 µm materials. For the analysis of large biomolecules the Accucore pore size has been further optimized and a C4 phase with reduced hydrophobic retention has been prepared. This 150 Å pore size enables the effective analysis of molecules unable to penetrate into smaller diameter pores and the low hydrophobicity C4 phase results in protein separation by hydrophobicity.

Method Details

Instrument parameters

Instrument Parameter	Value
Run Time Length	20.003 min
Wavelength 1	214.0 nm
Wavelength 2	281.0 nm
Mobile_Phase_A	0.1 % formic acid in water
Mobile_Phase_B	0.1 % formic acid in acetonitrile
Flow_Rate	300 nL/min
Backpressure_(100%_aqueous)	204 bar
Run_time	15 minutes + equillibration time
Column_Temperature	40 °C
Injection_details	0.25 µL 2 pmol/µL solution
UV_detector_wavelength	214 nm

Gradient Details

Ret. Time [min]	Flow [ml/min]	%B [%]	%C [%]	%D [%]
0.000	0.000	0.0	0.0	0.0
1.000	0.000	30.0	0.0	0.0
11.000	0.000	60.0	0.0	0.0
12.000	0.000	95.0	0.0	0.0
15.000	0.000	95.0	0.0	0.0

Column Details

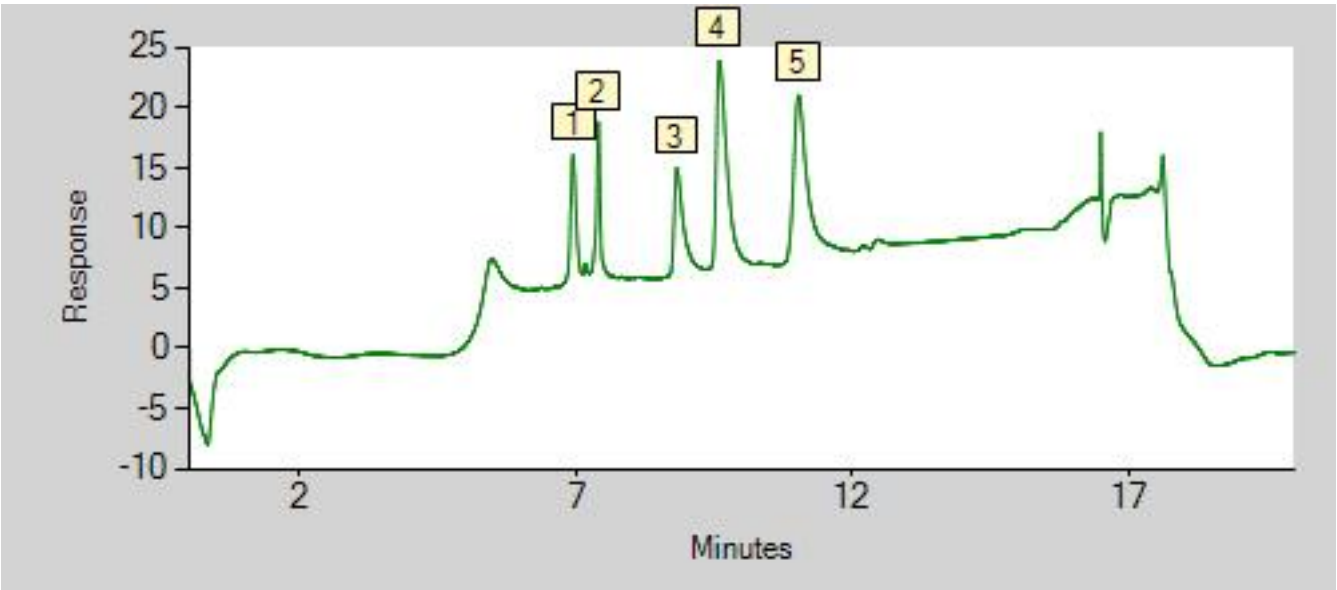
column_A	
Manufacturer	Thermo Fisher Scientific
Model	Accucore 150-C4
Diameter	75
Length	150
Particle Size	2.6
Packing Material	150-C4

System information

Instrument Type	UHPLC
UHPLC	Generic
UV Detector model	VWD-3400

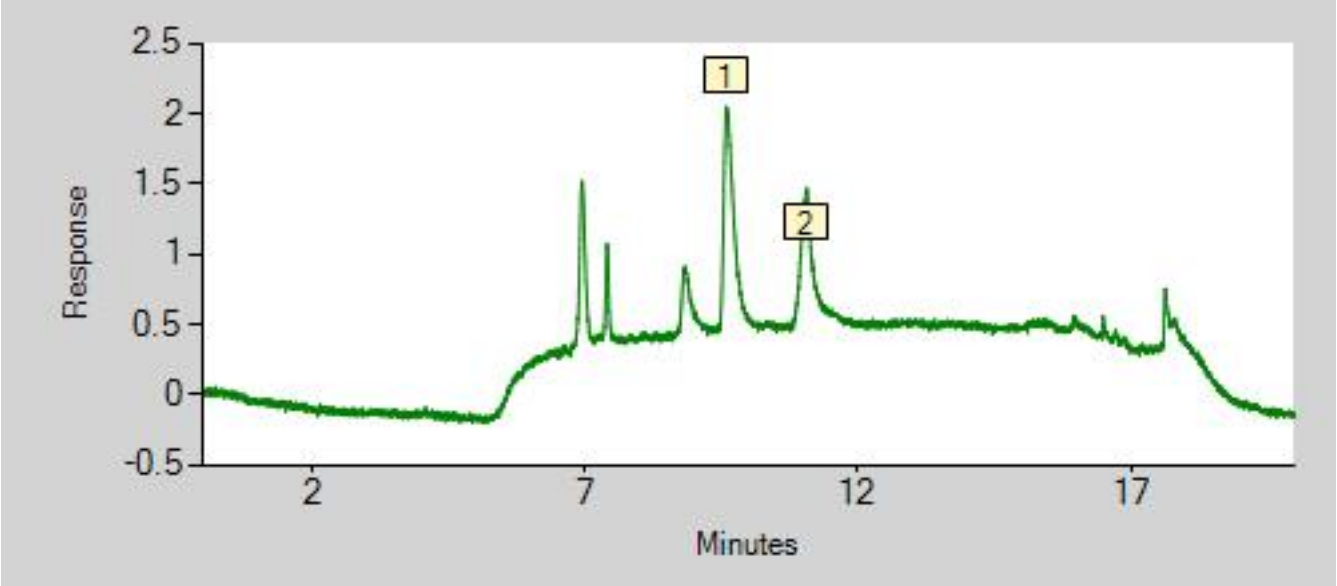
Results

Channel UV_VIS_1



No	Peak_Name	*Compound_Class	Retention_Time	Peak_Area	Peak_Area_pc	Peak_Height	Peak_Height_pc	Plates_(USP)	Resolution_(USP)	Tailing_Factor_(USP)
1	cytochrome C	Protein	6.940	1.143	9.95	10.162	16.29	24095	3.24	1.18
2	Insulin	Protein	7.403	1.008	8.78	12.712	20.38	76490	7.10	0.99
3	Myoglobin	Protein	8.823	1.720	14.98	8.855	14.19	14490	2.41	2.18
4	Carbonic Anhydrase	Protein	9.590	3.804	33.13	17.069	27.36	12507	3.86	2.24
5	Ovalbumin	Protein	11.023	3.808	33.16	13.590	21.78	12176	n.a.	1.81

Channel UV_VIS_2



No	Peak_Name	*Compound_Class	Retention_Time	Peak_Area	Peak_Area_pc	Peak_Height	Peak_Height_pc	Plates_(USP)	Resolution_(USP)	Tailing_Factor_(USP)
1	Carbonic Anhydrase	Protein	9.590	0.335	60.81	1.590	62.38	23079	4.34	2.03
2	Ovalbumin	Protein	11.053	0.216	39.19	0.959	37.62	11031	n.a.	1.12

Appendix

The application can be accessed at http://dlibrary.dionex.com/Public/View.aspx?ApplicationID=379

Available Downloads

Filename	Size(bytes)
Analysis of Intact Proteins.pdf	616672

Related Information

No related information available.