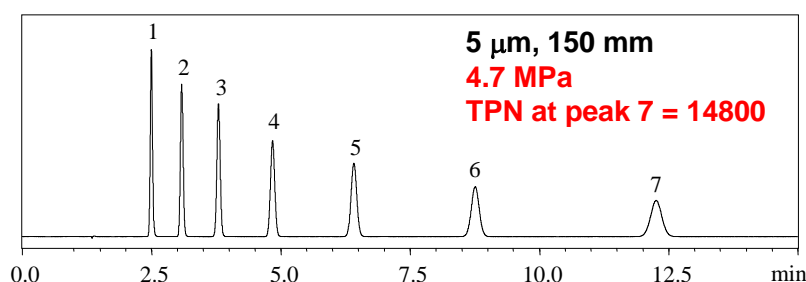


# Nexera Application Data Sheet No. 3

## The UHPLC with the Ultimate in Resolution

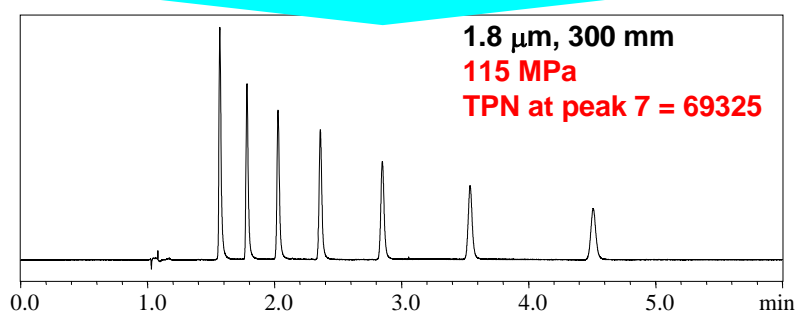
Nexera supports the use of long analytical columns thereby maximizing separation efficiency thanks to a high pressure limit of up to 130MPa. In this example, two 150 mm columns with 1.8  $\mu\text{m}$  particle size were connected sequentially thereby increasing Nexera's theoretical plate number to about 70,000.



Column : ODS (4.6 mm I.D. x 150 mm, 5  $\mu\text{m}$ )  
Mobile Phase: Water/Acetonitrile = 3/7  
Flow Rate : 1.0 mL/min  
Column Temp. : 40  $^{\circ}\text{C}$   
Detection : UV 245 nm

1. Acetophenone
2. Propiophenone
3. Butyrophenone
4. Valerophenone
5. Hexanophenone
6. Heptanophenone
7. Octanophenone

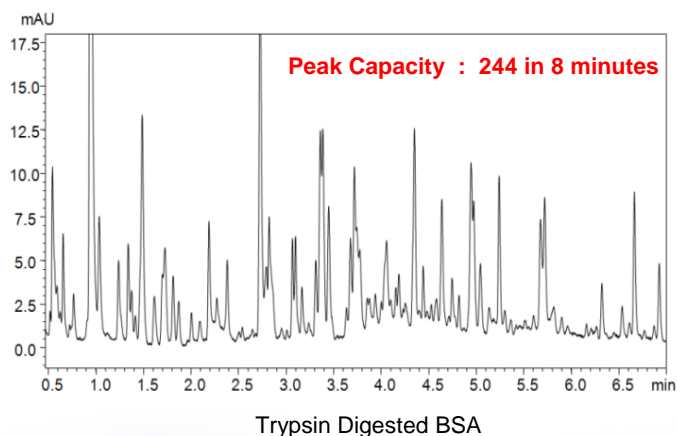
**5 times higher separation efficiency**



Column : ODS (2.1 mm I.D. x 150 mm x 2, 1.8  $\mu\text{m}$ )  
Mobile Phase: Water/Acetonitrile = 2/8  
Flow Rate : 0.5 mL/min  
Column Temp. : 50  $^{\circ}\text{C}$   
Detection : UV 245 nm

### Ultra-High Resolution Analysis of Trypsin-digested BSA

The peak capacity shows how many peaks can be separated during a given analysis time. For researchers it's a key factor in achieving high peak resolution of complex samples. The extremely small dead volume of Nexera insures high peak capacity of any analysis. As shown by a gradient elution of 1 pmol/ $\mu\text{L}$  of the BSA digested with Trypsin the peak capacity can reach 244 peaks in 8 minutes, or one peak every 2 seconds.



Column : ODS (2.1 mm I.D. x 100 mm, 1.8  $\mu\text{m}$ )  
Mobile Phase : A : 0.03% TFA in Water  
B : 0.03% TFA in Acetonitrile  
Gradient : B 5%  $\rightarrow$  40% (8 min)  
Flow Rate : 0.9 mL/min  
Column Temp. : 40  $^{\circ}\text{C}$   
Detection : UV 214 nm

The following formula was used to calculate peak capacity: Peak Capacity =  $\frac{t_g}{W} + 1$   
where  $t_g$  is gradient time and  $W$  stands for the USP peak width.