Raptor™ LC Columns
Selectivity Accelerated

General Applications

The Effects of LC Particle Choice on Column Performance: 2.7 vs. 5 µm Diameter Superficially Porous Particles (SPP)

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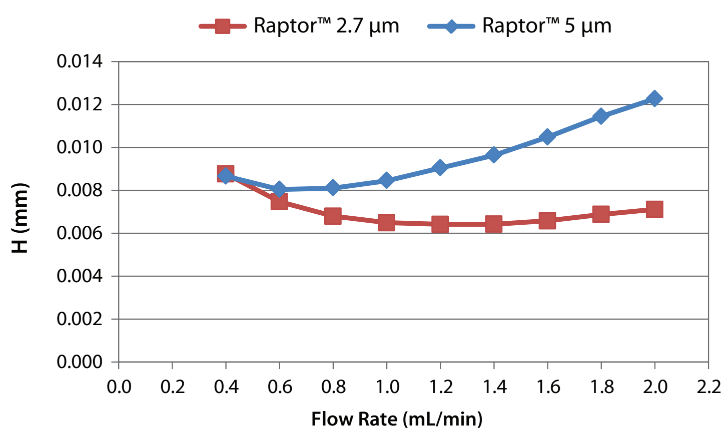
Superficially porous particles (commonly referred to as SPP or "core-shell" particles) have been proven to provide fast and efficient LC separations. These particles feature a solid, impermeable core enveloped by a thin, porous layer of silica that offers significantly higher efficiency and sensitivity than traditional fully porous particles. Restek's Raptor™ SPP LC columns are available in both 2.7 and 5 µm diameter particle sizes, giving analysts the flexibility to select the most appropriate size for their specific assay. However, the best LC particle choice may not always be clear. In this technical note, we will examine the differences in efficiency, sensitivity, and pressure between Raptor™ LC columns packed with 2.7 vs. 5 µm diameter particles and provide advice on making the appropriate particle choice based on the intended experimental conditions and instrument capability.

Efficiency

The relationship between column efficiency and linear velocity, or flow rate, can be illustrated using a van Deemter plot. Column efficiency is represented by plate height (H); the smaller the plate height at a given flow rate, the more efficient the column. The end result is sharper peaks and increased resolution. As shown in Figure 1, Raptor™ 2.7 µm columns display on average 25% more efficiency than Raptor™ 5 µm SPP columns across the flow rates tested. In addition, minimal loss in efficiency was observed at higher flow rates on the Raptor™ 2.7 µm column. For a 4.6 mm ID column, flow rates from 1.0 to 1.6 mL/min yielded the highest efficiency for our 2.7 µm diameter particle column; while flow rates ranging from 0.4 to 1.0 mL/min yielded the highest efficiency for our 5 µm diameter particle column.

Figure 1: Raptor™ 2.7 µm SPP columns maintain efficiency, even at elevated flow rates.

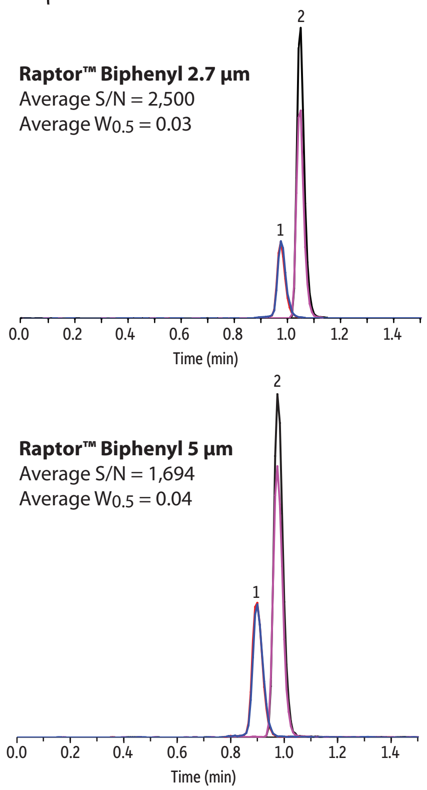


Column: Dimensions: 150 mm x 4.6 mm ID; Temp.: 30 °C;
Mobile Phase: Water:Acetonitrile (45:55); **Detection:** 254 nm;
Test Probes: Uracil and Biphenyl.

Sensitivity

Sensitivity can be measured by comparing signal-to-noise ratios (S/N) for a particular peak. Signal response can be increased by reducing peak width, thereby making peaks sharper and increasing sensitivity. Since superficially porous particles are less porous due to their solid, impermeable core, they offer a more direct diffusion path over fully porous particles, which results in reduced peak dispersion and narrower peaks. To demonstrate the impact of LC particle choice on S/N, a common pharmaceutical and its metabolite were analyzed on separate Raptor™ Biphenyl columns packed with 2.7 vs. 5 µm particles. The resulting chromatograms, peak widths, and S/N are compared in Figure 2. The Raptor™ 2.7 µm diameter particle column displays an average increase in S/N of 32% accompanied by a 25% decrease in average peak width when compared to the Raptor™ 5 µm diameter particle column.

Figure 2: Our 2.7 µm particles offer an average 32% increase in signal-to-noise ratio (i.e., greater sensitivity) over 5 µm.



Peaks	t_R 2.7 µm (min)	t_R 5 µm (min)	Precursor Ion	Product Ion 1	Product Ion 2
1. Naltrexone	0.98	0.90	342.4	270.1	267.1
2. 6-β-Naltrexol	1.06	0.98	344.4	308.0	254.1

Column
Temp.: 30 °C
Sample
Conc.: Prepared 500 ng/mL in human urine. Diluted 5x in mobile phase A. Final conc. = 100 ng/mL
Inj. Vol.: 10 µL
Mobile Phase
A: Water + 0.1% formic acid + 2 mM ammonium formate
B: Water:methanol (5:95) + 0.1% formic acid + 2 mM ammonium formate

Time (min)	Flow (mL/min)	%A	%B
0.00	0.5	70	30
1.50	0.5	0	100
1.51	0.5	70	30
3.00	0.5	70	30

Detector MS/MS
Ion Mode: ESI+
Instrument HPLC
Notes Top: Raptor™ Biphenyl 50 mm x 2.1 mm, 2.7 µm (cat.# 9309A52)
Bottom: Raptor™ Biphenyl 50 mm x 2.1 mm, 5 µm (cat.# 9309552)

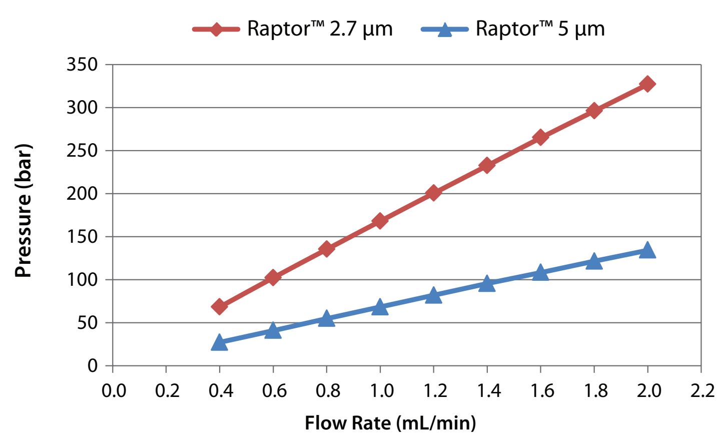
The S/N and $W_{0.5}$ calculations were made using Product Ion 1 (black trace).

LC_CF0597

Pressure

One of the primary advantages of SPP is its ability to provide increased column efficiency, often with similar or even reduced backpressure, when compared to fully porous particles. By decreasing the size of superficially porous particles, efficiency improves and pressure increases at a rate inversely proportional to the square of the particle size. In Figure 3, column backpressure is shown to increase by approximately 150% on average across the instrument flow rates tested (0.4 to 2 mL/min) when switching from a 5 µm diameter Raptor™ particle to a 2.7 µm diameter particle. Additional parameters that contribute to operating pressure include column dimensions, mobile phase composition, and sources of flow restriction on the LC and detection systems.

Figure 3: Switching from a 5 µm to 2.7 µm Raptor™ particle increases backpressure approximately 150%.



Column: Dimensions: 150 mm x 4.6 mm ID; Temp.: 30 °C; **Mobile Phase:** Water:Acetonitrile (45:55).

Conclusion

It is important to consider instrumentation and assay objectives when choosing between Raptor™ 2.7 vs. 5 µm diameter particle SPP LC columns.

Raptor™ 5 µm columns:

Raptor™ 5 µm diameter particle columns display low backpressure as well as good efficiency and sensitivity. These columns can be substituted into existing methods to increase analysis speed on traditional LC systems, especially those with pressure limitations. Raptor™ 5 µm SPP is an ideal LC particle choice for fast assays containing fewer analytes.

- Large amount of system volume.
- Maximum operating pressure of 400 bar.
- Fewer compounds requiring less peak capacity.

Raptor™ 2.7 µm columns:

Raptor™ 2.7 µm diameter particle columns exhibit greater efficiency and sensitivity than 5 µm SPP at the cost of higher pressures. Since extra-column effects are most pronounced on short, small-diameter columns packed with small particles, 2.7 µm columns are best suited for instrumentation with reduced system volume that can sustain pressures up to 600 bar. Raptor™ 2.7 µm SPP is the right LC particle choice for larger analyte lists that require additional peak capacity.

- Minimal system volume.
- Maximum operating pressure 600 bar.
- Large number of compounds requiring more peak capacity.

Lit. Cat.# GNAR2079-UNV

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