Stationary Phase: Silica

OH

Ο



**Selectivity Accelerated** 

# Raptor HILIC-Si: Simplify the Switch to HILIC

- Retain polar compounds without ion-pairing reagents.
- 2.7 μm Raptor core-shell particles provide the speed of SPP.
- Ideal for increasing sensitivity and selectivity in LC-MS analyses.
- Fully compatible with both HPLC and UHPLC.

## **Ready for HILIC?**

Check out our article on how to avoid common problems. www.restek.com/HILICtips



Pure Chromatography

www.restek.com/raptor

## The Raptor HILIC-Si Column

With Raptor LC columns, Restek chemists became the first to combine the speed of superficially porous particles (also known as SPP or "core-shell" particles) with the resolution of highly selective USLC technology. This new breed of chromatographic column allows you to more easily achieve peak separation and faster analysis times without expensive UHPLC instrumentation. Restek is extending the speed and reliability of Raptor to the HILIC realm with the addition of the Raptor HILIC-Si column.

Hydrophilic-interaction chromatography (HILIC) (1) is an increasingly popular alternative to reversed-phase LC for challenging polar analytes because it provides better retention of water-soluble compounds that are separated by polar differences (Figure 1). The Raptor HILIC-Si column simplifies the switch to HILIC because it delivers rug-ged Raptor performance, provides SPP column speed for faster analyses than traditional FPP silica columns, retains polar compounds without ion-pairing reagents, and is fully reliable, efficient, and selective with LC-MS compatible mobile phases.

### Order yours today at www.restek.com/raptor

Figure 1: Use HILIC when greater retention of polar analytes is needed. In HILIC mode, the aqueous mobile phase is the strong (or eluting) solvent versus the more familiar reversed-phase mode, where elution is the result of the organic solvent strength. HILIC Organic Mobile Phase Polar H<sub>2</sub>O Layer HILIC Phase (Hydrophilic) Metention Elution

#### **Reversed Phase**





(1) A.J. Alpert, Hydrophilic-interaction chromatography for the separation of peptides, nucleic acids and other polar compounds, J. Chromatogr. 499 (1990) 177–196.



### Raptor Technology Ensures Rugged, Reproducible Performance in the HILIC Realm

Raptor LC columns are well known for their rugged dependability, and the new Raptor HILIC-Si column brings the consistency of Raptor performance to HILIC. Lot to lot, column to column, and injection to injection, every Raptor HILIC-Si column gives a consistent performance that you can count on (Figures 2 and 3). Simplify your move to HILIC with the reliability of Raptor HILIC-Si columns. **Figure 2:** Raptor HILIC-Si columns maintain efficiency at any dimension, even at operating pressures up to 575 bar so you can run at high linear velocities with confidence.



**Figure 3:** Strict quality control guarantees that rugged Raptor HILIC-Si columns provide reproducible results lot after lot and injection after injection.



Column: Raptor HILIC-Si (cat.# 9310A52); Dimensions: 50 mm x 2.1 mm ID, Particle Size: 2.7 µm; Temp.: 30 °C; Sample: Diluent: Mobile phase A:mobile phase B (10:90); Conc.: 1 ng/mL; Inj. Vol.: 10 µL; Mobile Phase: A: Water, 100 mM ammonium formate, pH 3.0; B: Acetonitrile; Gradient (%B): 0.00 min (90% B), 5.00 (90% B); Flow: 0.3 mL/min; Detector: MS/MS; Ion Mode: ESI+; Mode: MRM; Instrument: UHPLC



## Raptor SPP + HILIC = More Speed, More Sensitivity

What makes Raptor HILIC-Si columns special? The answer is simple: you get the speed of a Raptor SPP column with the unique separating power of the HILIC retention mechanism. The benefits of superficially porous particles (SPP) are well known. SPP columns are characterized by a layer of porous silica bonded to a solid silica core, which gives faster, more efficient analyses. As shown in Figure 4, when you keep instrument parameters constant (flow, gradient, temperature) and compare a 3 µm fully porous particle (FPP) silica column to a 2.7 µm Raptor HILIC-Si SPP column, the benefits become clear. Raptor HILIC-Si columns combine faster analysis times with higher sensitivity so you can increase sample throughput and lower limits of quantification (LOQs) for difficult-to-retain polar analytes.





## Raptor HILIC-Si Performance Gains vs. RP: Say Good-bye to Ion-Pairing Reagents

HILIC methods are becoming more common as analysts search for better solutions to challenging reversed-phase (RP) analyses. But, when is HILIC a beneficial alternative to a standard RP approach? HILIC should be considered when analyzing small polar compounds that are difficult to retain in RP mode without the use of ion-pairing reagents in the mobile phase. For example, paraquat and diquat are highly charged quaternary amine herbicides that are often analyzed in RP mode using ion-pairing reagents (Figure 5). But, these reagents can contaminate your LC-MS/MS and require the system be taken off-line frequently for extensive cleaning. With Raptor HILIC-Si columns, ion-pairing reagents are not needed and paraquat and diquat are quickly retained and resolved with MS-friendly solvents and buffers so your instrument stays up and running longer (Figure 6).





## Raptor HILIC-Si Improves the Performance of Your LC-MS/MS

One of the most striking ways that HILIC analyses differ from standard RP analyses is that in HILIC methods the aqueous mobile phase is the stronger solvent. It provides differential selectivity that helps retain small polar analytes until after the sample matrix has eluted. In addition, the higher concentration organic mobile phases used in HILIC methods improve solvent evaporation during electrospray ionization, leading to increased sensitivity for LC-MS/MS methods. The following applications illustrate the decrease in matrix interference and increase in LC-MS/MS sensitivity that can be obtained using HILIC conditions and a Raptor HILIC-Si column.

#### Folic Acid and Metabolites

Folate deficiency is considered a risk factor for a wide range of human health problems including neural tube defects in newborns, cardiovascular diseases, Alzheimer's disease, and certain forms of cancer. The levels of folic acid and its metabolites in plasma are used as biomarkers to diagnose folate deficiency. However, when folic acid and its metabolites are extracted from plasma, phospholipids are also extracted and can cause matrix effects that make accurate quantitation difficult. Even with a good sample preparation protocol, 100% removal of phospholipids is difficult, and even at low levels they can interfere with the target analytes and also contaminate the MS source. By switching to a HILIC separation with a Raptor HILIC-Si column, you can quickly and easily separate the matrix components from the target analytes and accurately quantitate these important biomarkers for folate deficiency (Figure 7).



Column: Raptor HILIC-Si (cat.# 9310A5E); Dimensions: 50 mm x 3.0 mm ID, Particle Size: 2.7 µm; Temp.: 30 °C; Sample: Diluent: 20 mM Ammonium acetate in acetonitrile:water (80:20) containing 10 mg/mL 2-mercaptoethanol); Inj. Vol.: 5 µL; Mobile Phase: A: 50:50 Water:acetonitrile, 20 mM ammonium acetate; B: 20:80 Water:acetonitrile, 20 mM ammonium acetate; Gradient (%B): 0.00 min (100% B), 3.00 min (0% B), 3.20 min (0% B), 5.21 min (100% B), 5.21 min (100% B), Flow: 0.5 mL/min; Detector: MS/MS; Ion Mode: ESI+; Mode: MRM; Instrument: UHPLC; Notes: For sample preparation details, enter chromatogram LC\_CF0698 in the www.restek.com search.

#### Monoamine Neurotransmitters and Metabolites

Measurements of monoamine neurotransmitters and their metabolites in plasma and urine are commonly used for clinical diagnosis and monitoring of neuroblastoma and pheochromocytoma. Quantifying free metanephrine and normetanephrine is the most sensitive and accurate test for this purpose, but analysis of these polar compounds using reversed-phase LC is problematic due to very limited retention and poor sensitivity. As shown in Figure 8, these polar metabolites can be adequately retained on a Raptor HILIC-Si column and detected at 50 pg/mL in human plasma, providing the sensitivity needed for clinical purposes (Figure 8).





#### Food Adulterants

Foods that contain a high protein content command a higher price, which can result in the illegal practice of food adulteration using nitrogen-rich compounds, such as melamine, to make the protein content appear higher than the actual value. Due to its potential for toxicity, testing for melamine and other structurally related compounds is required in many countries for foods, feed materials, and pharmaceutical components. The method shown in Figure 9 provides excellent retention of these highly polar analytes with a separation time of only 3.5 minutes and a complete cycle time of just 8 minutes using a Raptor HILIC-Si column.





# **Raptor HILIC-Si:** Simplify the Switch to HILIC

#### Where's my 5 µm column?

Because HILIC methods use highly organic mobile phases, they generate very low backpressures. We've simplified your move to HILIC by offering the Raptor HILIC-Si column in a 2.7 µm particle size only. Our testing and applications development demonstrated that 2.7 µm columns offer higher efficiency than 5 µm columns, and they are compatible with any HPLC or UHPLC instrument you have in your lab.

#### Raptor HILIC-Si LC Columns (USP L3)

RESTER			<b>BEZTEK</b>	
Length	2.1 mm cat.#	3.0 mm cat.#	4.6 mm cat.#	
2.7 µm Columns				
30 mm	9310A32			
50 mm	9310A52	9310A5E	9310A55	
100 mm	9310A12	9310A1E	9310A15	
150 mm	9310A62	9310A6E	9310A65	

#### **EXP Reusable Fittings for HPLC & UHPLC**

for 10-32 fittings and 1/16" tubing

Effortlessly achieve 8,700+ psi HPLC seals by hand! (Wrench tighten to 20,000+ psi.) Hybrid titanium/PEEK seal can be installed repeatedly without compromising your seal.



Description	qty.	cat.#
EXP Hand-Tight Fitting (Nut w/Ferrule)	ea.	25937
EXP Hand-Tight Fitting (Nut w/Ferrule)	10-pk.	25938
EXP Hand-Tight Nut (w/o Ferrule)	ea.	25939

Hybrid Ferrule U.S. Patent No. 8201854, Optimize Technologies. Optimize Technologies EXP Holders are Patent Pending. Other U.S. and Foreign Patents Pending. The Opti- prefix is a registered trademark of Optimize Technologies, Inc.

#### **Raptor EXP Guard Cartridges**



Protect your investment and extend the life of our already-rugged LC columns and change guard column cartridges by hand without breaking fluid connections—no tools needed!

#### **EXP Direct Connect Holder**

Description	qty.	cat.#
EXP Direct Connect Holder for EXP Guard Cartridges (includes hex-head fitting & 2 ferrules)	ea.	25808
Maximum holder pressure: 20,000 psi (1,400 bar)		

#### **Raptor EXP Guard Column Cartridges**

	Particle		5 x 2.1 mm	5 x 3.0 mm	5 x 4.6 mm
Description	Size	qty.	cat.#	cat.#	cat.#
Raptor HILIC-Si EXP Guard Column Cartridge	2.7 µm	3-pk.	9310A0252	9310A0253	9310A0250

Maximum cartridge pressure: 600 bar/8,700 psi (2.7 µm) or 400 bar/5,800 psi (5 µm) Raptor SPP LC columns combine the speed of SPP with the resolution of USLC technology.

Learn more at www.restek.com/raptor

### Experience Selectivity Accelerated. Order the Raptor HILIC-Si today at www.restek.com/raptor

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