SunFire™ C_{18}: A Unique RPLC Stationary Phase for Higher Loading and Improved Peak Shapes

Fang Xia, Jie Y. Cavanaugh and Diane M. Diehl, Waters Corporation, Milford, Massachusetts, USA.

The new SunFire™ C_{18} columns are reversed-phase HPLC columns designed for high preparative mass loading, excellent efficiency and peak shapes and stability under low pH conditions.

Today's chromatographers often struggle with tailing peaks that limit resolution and mass loading. High silanol activity contributes significantly to peak tailing in reversed-phase liquid chromatography (RPLC) columns. Other factors, such as the purity of the silica, are also keys to reducing peak tailing. SunFire™ C_{18} columns are engineered with highly pure raw materials and a tightly controlled synthesis process. They provide high efficiencies and symmetric peak shapes for the analysis of acids, neutrals and bases and they exhibit superior lifetimes under low pH conditions. The SunFire™ C_{18} preparative columns are manufactured with the patent pending OBD™ design to ensure ease of scale-up as well as the same efficiency, stability and reliability as the analytical columns.

Experimental Conditions

Scale-up Conditions:

Columns: SunFire™ C_{18} 4.6 × 50 mm, 5 µm and 19 × 50 mm, 5 µm
Mobile phase A: 0.1% trifluoroacetic acid in water
Mobile phase B: 0.1% trifluoroacetic acid in acetonitrile
Flow-rate: 1.4 mL/min analytical, 23.9 mL/min preparative
Analytical gradient: 5 min linear from 5% to 30% B, with 1 min initial hold time
Preparative gradient: 5 min linear from 5% to 30% B with 1.79 min initial hold time
Injection volume: 23 µL (analytical) and 400 µL (preparative)
Sample mixture: nadolol (100 mg/mL), metoprolol (100 mg/mL), and propranolol (50 mg/mL) prepared in DMSO
Detection: UV at 270 nm

Loadability Conditions:

Column dimensions: 4.6 × 150 mm, 5 µm (all silica-based C_{18})
Mobile phase A: 0.1% trifluoroacetic acid in water
Mobile phase B: 0.1% trifluoroacetic acid in acetonitrile
Flow-rate: 1.0 mL/min analytical
Gradient: 15 min linear from 20% to 85% B, with 2 min initial hold time
Injection volume: 10 µL
Sample mixture: ketoconazole (25 mg/mL), econazole (50 mg/mL), and miconazole (50 mg/mL) prepared in DMSO
Mass loading: 1250 µg
Detection: UV at 254 nm
Instrument: Waters® AutoPurification™ System

Results

The retention and separation of the three β-blockers on the analytical column is shown in Figure 1(a). The total load is

Figure 1: Scale-up of the ß-blockers separation from analytical to preparative SunFire™ C_{18} columns. (a) SunFire™ C_{18}, 4.6 × 50 mm, 5 µm and (b) SunFire™ C_{18} Prep, 19 × 50 mm, 5 µm. Analytes: 1 = nadolol, 2 = metoprolol, 3 = propranolol.

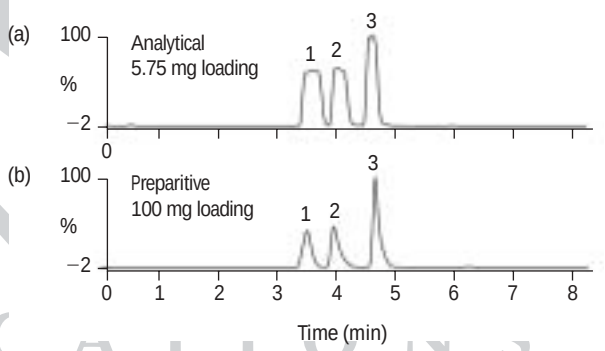
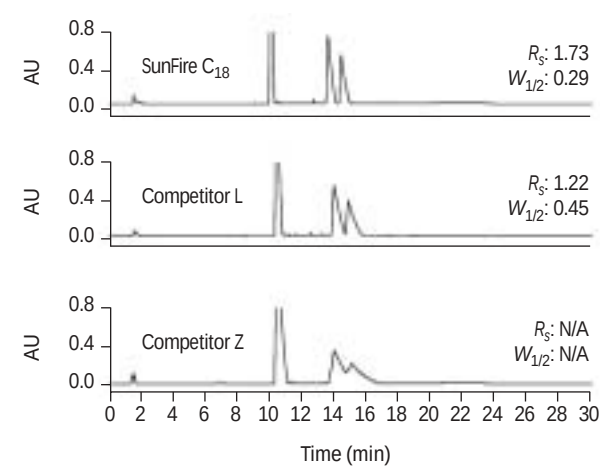


Figure 2: Separation of basic antifungal drugs on SunFire™ C_{18} column compared to two competitors' silica-based C_{18} columns. Analytes in order of elution: ketoconazole, econazole and miconazole. Resolution (R_s) values are calculated based on the last two peaks. $W_{1/2}$ values are calculated based on the width at half-height of the last peak.



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Mass loading: 1250 µg
Detection: UV at 254 nm
Instrument: Waters® AutoPurification™ System

Results

The retention and separation of the three β-blockers on the analytical column is shown in Figure 1(a). The total load is 5.75 mg, and the flattened profiles reflect the saturation of the PDA detector. The mass load was proportionally scaled-up and run on the preparative column as shown in Figure 1(b). Note the direct scale up, excellent peak shapes and total mass load of 100 mg.

The separation and loadability of the three antifungal drugs on SunFire™ C_{18} and two competitive silica-based C_{18} columns is shown in Figure 2. Under the same high mass loading, only SunFire™ C_{18} columns provide baseline resolution and excellent peak shapes for this separation.

Conclusions

New SunFire™ C_{18} columns provide excellent efficiency and peak shape, high mass loading, and ease of scale-up.

Synthetic peptide separation

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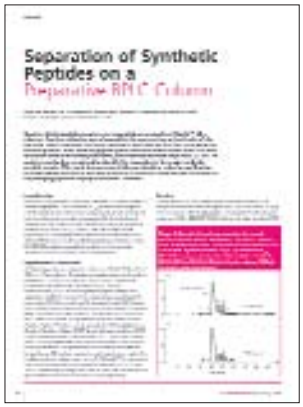

From Waters comes an application note describing separation of synthetic peptides on a preparative RPLC column. Crude synthetic peptide samples were separated on preparative Atlantis™ dC_{18} columns. Fraction collection was triggered by the mass-to-charge (*m/z*) ratio of the peptides, which specified the target peptide in the collected fraction to increase the fraction purities. The study demonstrates high mass loading, selective purification, high recoveries and purity, and the capability of using mass-directed fractionation for purifying peptides on preparative RPLC columns.
Waters Corp., Milford, Massachusetts, USA.

Reversed-phase HPLC

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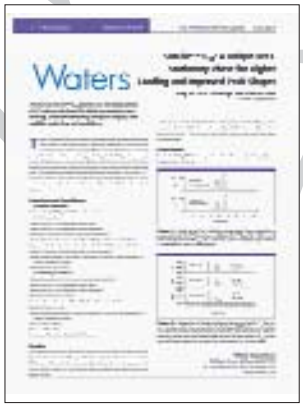

From Waters comes an application note highlighting how its SunFire™ C_{18} column reportedly provides excellent efficiency and peak shapes, high preparative mass loading and ease of scale-up. The note describes the retention and separation of three antifungal β-blockers on the Sunfire column and two competitive silica-based C_{18} columns. The preparative columns are manufactured with the patent pending OBD™ design to ensure equal efficiency, stability and reliability as the analytical columns.
Waters Corp., Milford, Massachussetts, USA.

Waters Corporation
34 Maple Street, Milford,
Massachussetts 01757, USA.
tel. +1 508 478 2000 **fax.** +1 508 478 1990
e-mail: info@waters.com **website:** www.waters.com