

Waters

Isolation of Impurities in Drugs using SunFire™ Columns

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SunFire™ C₁₈ analytical and OBD™ preparative columns are designed to provide maximum loadability in simple mobile-phase conditions, accurate scalability, and high peak capacity. They are ideal for isolating impurities in crude drug samples.

Nimodipine is a calcium-channel blocker, which acts on blood and heart vessels. Several impurities may be formed during its synthesis. The existence of impurities even in small amounts may influence the efficacy and safety of drugs. Identification of impurities is important to get insight of pharmaceuticals. Advanced analytical technologies, such as HPLC and MS-MS, are efficient ways to detect those trace impurities.

SunFire™ C₁₈ columns are engineered with highly pure raw materials and a tightly controlled synthesis process. These columns provide high efficiencies, maximum loading, and symmetric peak shapes for the analysis of acids, neutrals, and bases. SunFire™ C₁₈ preparative columns are manufactured with the Optimum Bed Density (OBD™) design to deliver consistent column-to-column performance, unmatched column lifetime with DMSO sample diluents, and accurate scalability.

Experimental Conditions

HPLC Conditions

Columns: SunFire™ C₁₈ 4.6 × 100 mm, 5 μm and 19 × 100 mm, 5 μm

Mobile Phase A: 0.1% formic acid in water

Mobile Phase B: 0.1% formic acid in acetonitrile

Flow Rate: 1.4 mL/min analytical, 23.9 mL/min preparative

Analytical Gradient: 10-min linear from 30% to 90% B, with 1 min initial hold time

Preparative Gradient: 10-min linear from 30% to 90% B with 1.97 min initial hold time

Injection Volume: 200 μL (analytical) and 3400 μL (preparative)

Sample: Crude nimodipine prepared in DMSO at 30 mg/mL

Total Mass Loading: 6 mg (analytical), 102 mg (preparative)

Detection: UV at 290 nm

Instrument: Waters® AutoPurification™ System

MS Conditions

Interface: Positive-mode ESI

Capillary: 3.5 kV

Cone: 30 V

Source Temperature: 150 °C

Desolvation Temperature: 250 °C

Cone Gas Flow: 50 L/h

Desolvation Gas Flow: 250 L/h

Instrument: Waters® Micromass Quattro micro™ API

Results

The separation of nimodipine and four impurities on the analytical column is shown in Figure 1A. The total load is 6 mg. The chromatograms are baseline enhanced in order to identify the impurity peaks. As shown in the figure, closely eluting impurity 2 has been detected and separated from nimodipine. The separation was proportionally scaled-up and run on the preparative column as shown in Figure 1B. Note the direct scale up, excellent peak shapes, and total mass load of 102 mg. Each peak in Figure 1A was collected in separate vials first, and then analyzed using MS-MS. Nimodipine has the molecular weight of 418.1 (peak N). The daughter ion scan of 419 was shown on Figure 2, top one. Similarly, the daughter ion scan was applied to get further structural information on peaks 1, 2, 3, and 4. Finally, we determined the structures based on the MS-MS data as shown in Figure 2.

Conclusions

Highly efficient isolation and direct scale-up are observed on SunFire™ C₁₈ analytical and preparative columns. This column technology ensures rapid target purifications with minimal chromatographic development. The combination of Waters' chemistry and triple-quadrupole mass spectrometers is a powerful tool for drug impurity profiling.

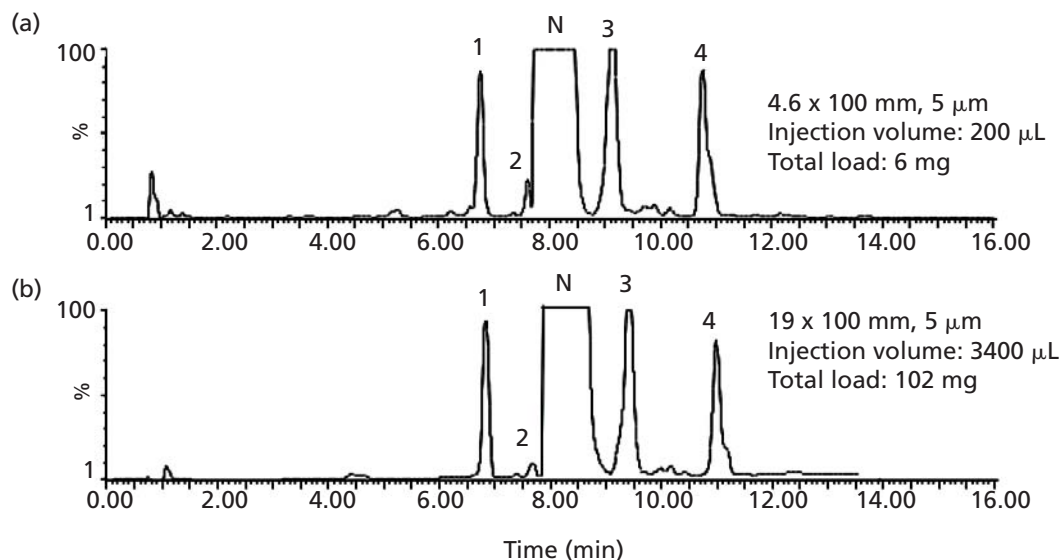


Figure 1: Scale-up of the separation of nimodipine and four impurities from analytical to preparative SunFire™ C₁₈ columns. A) SunFire™ C₁₈, 4.6 \times 100 mm, 5 μ m. B) SunFire™ C₁₈, OBD™, 19 \times 100 mm, 5 μ m. Peak annotation: Nimodipine (N) and its impurities (1-4).

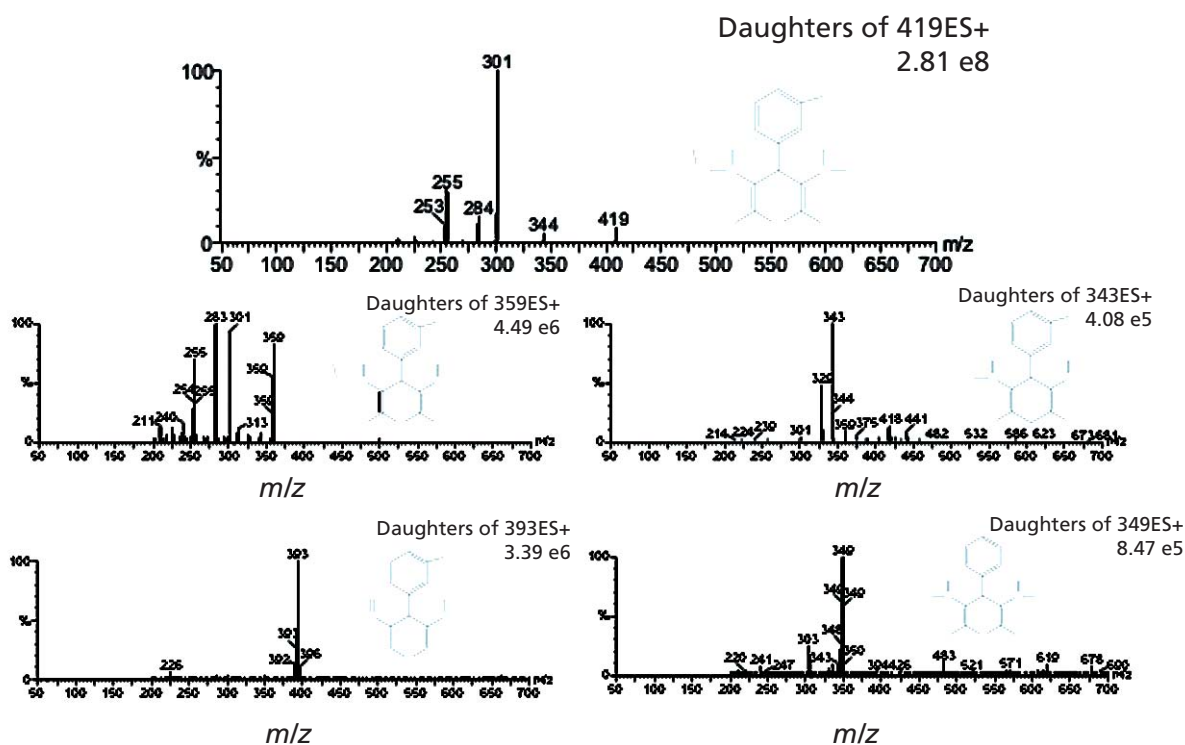


Figure 2: The mass spectrum and proposed structure of each impurity.