

An Improved Fast RP-HPLC Method for Rosuvastatin and its Degradation Products Using a Thermo Scientific Accucore PFP HPLC Column

Anirudha Rakhe, Tushar N. Mehta,
Centre of Excellence for Asia Pacific Laboratory, Thermo Fisher Scientific, Ahmedabad, India

Key Words

Rosuvastatin, core enhanced technology, Accucore PFP HPLC column, anti isomer separation, HPLC

Abstract

This application note demonstrates the advantages of using an Accucore™ PFP HPLC Column for the determination of rosuvastatin and its degradation products by HPLC-UV.

Introduction

Rosuvastatin is classified as a statin, a type of agent that inhibits cholesterol production in the liver. Rosuvastatin calcium is a synthetic lipid-lowering agent approved as a treatment for hypercholesterolemia.

Rosuvastatin is an inhibitor of HMG-CoA reductase, an enzyme that catalyzes the conversion of HMG-CoA to mevalonate, an early and rate-limiting step in cholesterol biosynthesis.

Rosuvastatin reduces cholesterol by increasing the number of low-density lipoprotein (LDL) receptors on the cell-surface to enhance uptake and catabolism of LDL. It also inhibits hepatic synthesis of hepatic very-low-density lipoprotein (VLDL), which reduces the total number of VLDL and LDL particles. The treatment reduces tri-glycerides (TG) and produces increases in high-density lipoprotein cholesterol (HDL-C.)

Accucore HPLC columns use Core Enhanced Technology™ to facilitate fast and high efficiency separations. The 2.6 µm diameter particles are not totally porous, but rather have a solid core and a porous outer layer. The optimized phase bonding creates a series of high coverage, robust phases. Accucore PFP uses an optimized perfluorinated benzene ring for more effective coverage of the silica surface. This coverage results in a significant reduction in secondary interactions and thus highly efficient peaks with very low tailing. The tightly controlled 2.6 µm diameter of Accucore particles results in much lower backpressures than typically seen with sub-2 µm materials.



This application note demonstrates the successful resolution between rosuvastatin and its anti isomeric impurity using an Accucore 2.6 µm column as compared to UHPLC¹, without the high backpressures associated with the technique.

Experimental Details

Consumables	Part Number
Water, from TKA Water Purification System	
Fisher Scientific HPLC grade potassium dihydrogen orthophosphate	13415
Fisher Scientific HPLC grade orthophosphoric acid	ZA260-500
Fisher Scientific HPLC grade acetonitrile	26892-1000
Fisher Scientific HPLC grade methanol	ZA452SK-4
Thermo Scientific Screw Top 2 mL Vial Kit w/PTFE/Silicone Closure	60180-600
Rosuvastatin procured from Customer	

Solutions

Working standard contained 1000 µg/mL of in mobile phase.

Separation Conditions	Part Number
Instrumentation:	Thermo Scientific Dionex UHPLC Plus Focused (Ultimate 3000) system
Column:	Accucore PFP 2.6 µm, 100 x 3.0 mm 17426-103030
Buffer preparation:	weigh 2.74 g of KH ₂ PO ₄ in 1000 ml of water and adjust pH-2.5 with o-phosphoric acid
Mobile phase:	70:29:1 (v/v) buffer:acetonitrile:methanol
Diluent:	50:50 (v/v) acetonitrile:water
Flow rate:	0.7 mL/minute
Column temperature:	30 °C
Injection details:	2.5 µL partial loop
Injection wash solvent:	50:50 (v/v) water:acetonitrile
UV detector wavelength:	243 nm
Backpressure:	approximately 230 bar

Results

The analysis was performed on an Accucore PFP 2.6 µm, 100 x 3.0 mm column - see Figure 1, which shows the result for acid degradation. Table 1 shows the results from six replicate injections at the 1 ppm level.

Parameter	Rosuvastatin
Retention time (minutes)	7.14
%RSD on retention time	0.09
Area	827.51
%RSD on area	1.08

Table 1: Retention time and area results for rosuvastatin

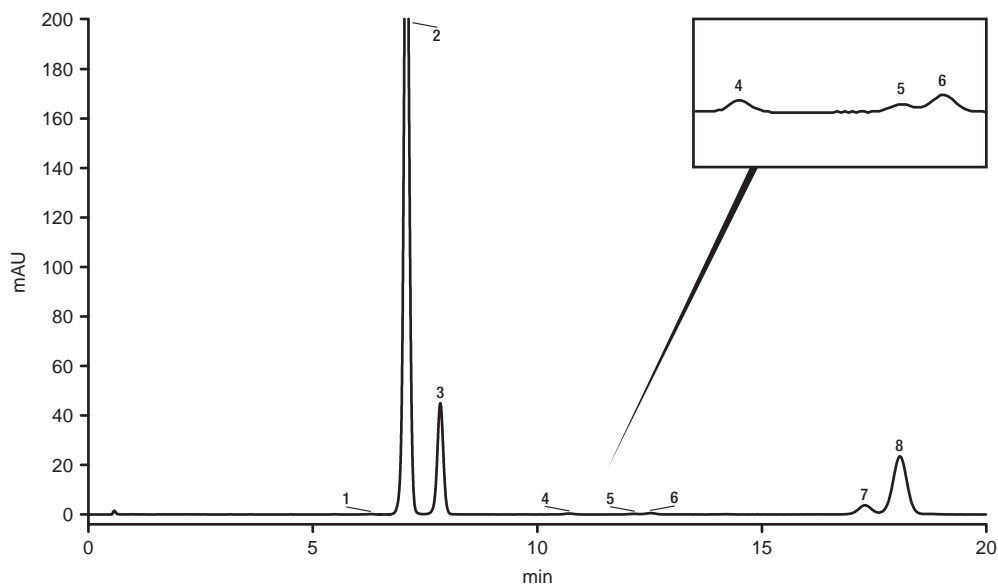


Figure 1: Chromatogram representing resolution of acid degradation for rosuvastatin

1. unknown impurity-1 2. rosuvastatin 3. anti isomer 4. unknown Impurity-2 5. unknown impurity-3
6. unknown impurity-4 7. unknown impurity-5 8. lactone impurity

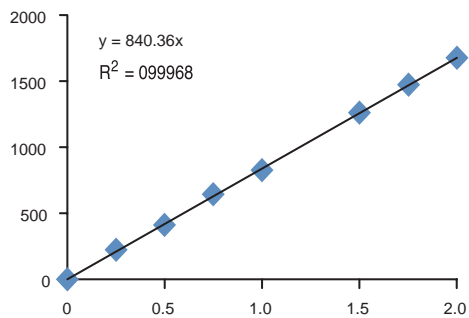


Figure 2: Linearity also established from LOQ (0.25 ppm) to 2 ppm. The value of R2 was found 0.99968

In documented degradation studies of rosuvastatin the acid degradation study showed more degradation than base or oxidation¹. Acid degradation was carried out using 1N HCl for which results are shown below in Table 2.

Peak name	t_r (Min)	% Rel. Area Response	Resolution
Rosuvastatin	6.98	75.06	N/A
Anti Isomer of Rosuvastatin	7.73	7.72	3.3
Lactone	18.11	15.64	27.7

Table 2: Representation for the extent of degradation and resolution of degradation products with acid induced degraded sample

The method in this application note has a shorter run time than the HPLC method in the literature². It also has comparable resolution of anti-isomer with reported UHPLC methods¹. Accucore HPLC columns provide benefits in terms of lower backpressures and reduced run times, when compared to the literature methods.

Replicate injections of rosuvastatin showed that Accucore PFP produced stable and reproducible results.

Conclusion

This application note shows that Accucore PFP is an excellent choice of column for resolution of anti isomeric impurities, demonstrating UHPLC like performance at HPLC backpressures¹.

References

1. Development and Validation of a Stability-Indicating UPLC Method for Rosuvastatin and its Related Impurities in Pharmaceutical Dosage Forms, *Quim. Nova*, 2011, Vol. 34, No. 2, 250-255.
2. Determination of Rosuvastatin in the Presence of its Degradation Products by a Stability-Indicating LC Method, Mehta, T. N.; Patel, A. K.; Kulkarni, G.; Subbaiah, G.; *J. AOAC Int.* 2005, 88, 1142.

thermoscientific.com/accucore

© 2012 Thermo Fisher Scientific Inc. All rights reserved. All trademarks are the property of Thermo Fisher Scientific Inc. and its subsidiaries. Specifications, terms and pricing are subject to change. Not all products are available in all countries. Please consult your local sales representative for details.

Australia 1 300 735 292 (free call domestic)
China +86 21 68654588 +86 10 84193588
+86 20 83145199 800 810 5118
France +33 (0)1 60 92 48 34
Germany +49 (0) 2423 9431 20 or 21

India +91 22 6742 9494 +91 27 1766 2352
Japan +81 3 5826 1615
New Zealand 0800 933 966 (free call domestic)
United Kingdom +44 (0)1928 534110
USA and Canada +1 800 332 3331
All Other Enquiries +44 (0) 1928 534 050

Technical Support
North America +1 800 332 3331
Outside North America +44 (0) 1928 534 440

Thermo
SCIENTIFIC

Part of Thermo Fisher Scientific