Case study | 001061

HPLC

Avoiding headaches by a straightforward analytical strategy for successful method transfer

Migrating USP Atorvastatin Calcium impurity analysis from an Agilent 1260 Infinity system to a Vanquish Flex UHPLC system

Authors

Nolan Dean[1], Sebastien Morin[2]

[1]Thermo Fisher Scientific, Greenville, NC, USA

[2]Thermo Fisher Scientific, Mississauga, ON, Canada

Keywords

HPLC method transfer protocol, HPLC method transfer guide, HPLC method transfer tips, HPLC impurity analysis method, atorvastatin

Application benefits

- Simplified method transfer from an Agilent™ 1260 Infinity™ system to a Thermo Scientific™ Vanquish™ Flex UHPLC system within USP guidelines
- Improved signal-to-noise using a Thermo Scientific™ Vanquish™ Diode Array Detector (DAD) HL

Goal

To demonstrate the simple transfer of an analytical HPLC method from an Agilent 1260 Infinity system to a Vanquish Flex UHPLC system

Introduction

Analytical method transfer is defined as the documented process that qualifies a laboratory (the receiving laboratory) to use an analytical method that originated in another laboratory (the transferring laboratory), whether that is internal or external to the receiving laboratory.[1] Even if current Good Manufacturing Practices (cGMP) require method lifecycle management, analytical method transfer and method modernization can be a barrier to upgrading to the latest technologies

patheon
by Thermo Fisher Scientific

thermo scientific

Revalidating an existing method while continuing to meet regulatory guidance can be a challenging and time-consuming endeavor. However, there are instances in which method transfer is essential, for example when the receiving laboratory does not have access to the same instrumentation on which the method was developed or when the initial system on which the method was developed is no longer fit for purpose. In these instances, there are a few strategies that can be put in place to avoid full revalidation and qualification, which have been discussed in a previous case study.[2] The following case study demonstrates one scenario that addresses some of the concerns typically associated with method transfer.

Atorvastatin, sold under the brand name Lipitor™, was one of the most commonly prescribed drugs in the United States in 2019.[3] The main therapeutic usage of this medication is to prevent cardiovascular disease in those at high risk and to treat abnormal lipid levels.[4] It is on the World Health Organization's List of Essential Medicines and was one of the top 10 blockbuster generic drugs in 2019.[5] It is thus a pharmaceutical drug that is produced in high volume and across multiple manufacturing sites. For these reasons, several generic companies and Contract Development and Manufacturing Organizations (CDMOs) must either develop novel analytical methods or transfer analytical methods from partner laboratories. In this specific example, we will demonstrate a strategy that was employed to transfer the USP Atorvastatin Calcium impurity analysis.

In our case, the method was originally validated on an Agilent 1260 Infinity Binary LC system. There were no absolute retention time requirements in the quality agreement, and the originating lab SOP did not require an official qualification study to qualify the novel instrument. For this matter, we applied the strategy below to transfer the method onto a Vanquish Flex UHPLC system:

- Perform system suitability (SST) on an already approved instrument and the new instrument using the same analyst, solutions, and column.
- Verify chromatography is consistent prior to proceeding with sample analyses (same relative retention time, no unexpected peaks, etc.).
- Verify sample results are consistent between systems.

Even though our laboratory had access to an Agilent 1260 Infinity system of similar configuration as the one used in the originator method, we decided to transfer the method onto the Vanquish Flex UHPLC system as it can lead to better performance in our experience. Additionally, since Atorvastatin Calcium impurity analysis is a readily available USP method,[6] the method transfer included allowable USP modifications.

Experimental

Materials

- Water, HPLC grade, 18.2 MΩ·cm resistivity or higher
- Ammonium acetate, HPLC grade, Fisher Scientific™ (Fisher Catalog # A639)
- Glacial acetic acid, ACS grade, Fisher Scientific™ (Fisher Catalog # A38C)
- Acetonitrile (ACN), Optima™ LC/MS grade, Fisher Scientific™ (Fisher Catalog # A995)
- Tetrahydrofuran (THF), HPLC grade 99.8%, unstabilized, Acros™ (Fisher Catalog # AC26829)
- *N,N*-Dimethylformamide, ACS grade, Fisher Scientific™ (Fisher Catalog # D119)
- Atorvastatin calcium, Certified, Supelco™ (Sigma-Aldrich Catalog # PHR1422)
- Atorvastatin related compound A, Certified, Supelco™ (Sigma-Aldrich Catalog # PHR1868)
- Atorvastatin related compound B, Certified, Supelco™ (Sigma-Aldrich Catalog # PHR1869)
- Atorvastatin related compound C, Certified, Supelco™ (Sigma-Aldrich Catalog # PHR1870)
- Atorvastatin related compound D, Certified, Supelco™ (Sigma-Aldrich Catalog # PHR1871)

Sample preparation

Atorvastatin calcium was prepared at 1 mg/mL in alternative diluent (ACN:stabilizer-free THF:water 1:1:2 v:v:v) with sonication to aid in dissolution.

Instrumentation and HPLC conditions

The instruments and the HPLC conditions used in this study are listed in Tables 1 and 2.

Data processing and software

Thermo Scientific™ Standard Instrument Integration (SII version 1.1) for Waters™ Empower™ 3 Chromatography Data System with Feature Release 4 was used.

Table 1. Instruments

	Agilent 1260 Infinity Binary LC	Vanquish Flex Quaternary UHPLC
Pump	• 1260 Bin Pump, G1312B • 1260 HiP Degasser, G4225A	• Quaternary Pump F, VF-P20-A
Sampler	• 1260 HiP ALS, G1367E: • 1290 Thermostat, G1330B Configured for 100 µL syringe injection	• Split Sampler FT, VF-A10-A, Configured for 100 µL max. injection volume (6850.1913 Sample loop, left, 100 µL, biocomp., VH/VF-A10, VH/VF-A40)
Column Compartment	• 1260 TCC, G1316A	• Vanquish Column Compartment, VH-C10-A-03 With Active Preheater, 6732.0110
Detector	• 260 DAD, G4212B With Max-Light 10 mm Cartridge Cell, G4212-60008	• Diode Array Detector HL, VH-D10-A With LightPipe 10 mm Flow Cell, 6083.0100B
System Base		• Vanquish Horizon/Flex, VF-S01-A-02

Table 2. HPLC conditions

Parameter	Value
Column	USP L7 C8, 4.6 × 250 mm, 5 µm
Solvent A	ACN, stabilizer free THF, and 3.9 g/L ammonium acetate in water (21:12:67 v:v:v)
Solvent B	ACN, stabilizer free THF, and 3.9 g/L ammonium acetate in water (61:12:27 v:v:v)
Diluent[1]	1: *N,N*-Dimethylformamide 2: ACN, stabilizer free THF, and water (1:1:2 v:v:v)
Flow rate	1.5 mL/min
Gradient[2]	Time (min) Mobile phase B % 0 6 40 6 70 80 85 100 100 100 105 0 115 0
Column temperature	35 °C
Autosampler temperature	Ambient
Detection	UV 244 nm, Bandwidth 4 nm, 2.0 Hz
Injection volume	20 µL
Needle wash	ACN:water (1:1 v:v)

[1]Diluent-1 provided unacceptable fronting and Diluent-2 was used per USP monograph

[2]As allowed per USP monograph and USP <621>, initial gradient conditions modified to 0 min 6% B to achieve a retention time of 26–34 min for the atorvastatin peak.

Results and discussion

As mentioned above, the method was evaluated on an Agilent 1260 Infinity system and a Vanquish Flex UHPLC system. Although there are no signal-to-noise requirements in the monograph, the ICH recommends a 10/1 ratio or greater to be acceptable for known peaks. Analysis of system suitability was performed with the Vanquish Flex UHPLC system and the Agilent 1260 system with the configuration indicated in Table 1 and using the same solutions and instrument conditions as described in Table 2. Both systems met the system suitability requirements (resolution NLT (no less than) 1.5) of the USP monograph, and both instruments presented similar peak shapes and chromatographic profiles. Therefore, the Vanquish Flex UHPLC system was deemed a suitable alternative for running this method. As seen in Figures 1 and 2, the systems exhibited differences in retention time. If required, and according to USP Monograph and USP General Chapter <621>, the initial gradient conditions may be adjusted to ensure matching retention time system to system. However, it is important to point out that unless stated in an SOP or quality agreement, exact retention time repeatability across instruments is not required from a regulatory standpoint as relative retention time and meeting the system suitability criteria are acceptable.

One particular improvement using the Vanquish Flex UHPLC system is an increased sensitivity with noise reduced by a factor of 2 compared to that of the Agilent 1260 system. The Vanquish Flex pump design reduces pressure and flow fluctuation caused by the pump, resulting in smoother baseline. This, combined with the Thermo Scientific™ Vanquish™ LightPipe™ flow cell, reduces noise considerably and so improves the sensitivity.

Improved instrument sensitivity can be vital for early detection of unknown and degradation impurities, which may be caused by manufacturing process deviation, storage, shipment, or further processing of the drug material. The results from this comparative study are summarized in Table 3.

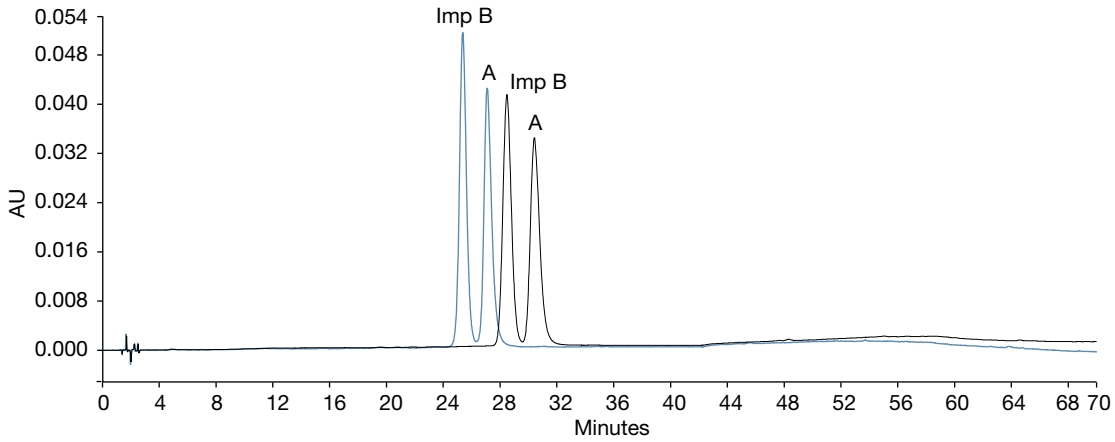

Figure 1. System suitability (Atorvastatin (A) and impurity B (Imp B)) standard solutions on an Agilent 1260 system (blue) and a Vanquish Flex UHPLC system (black)

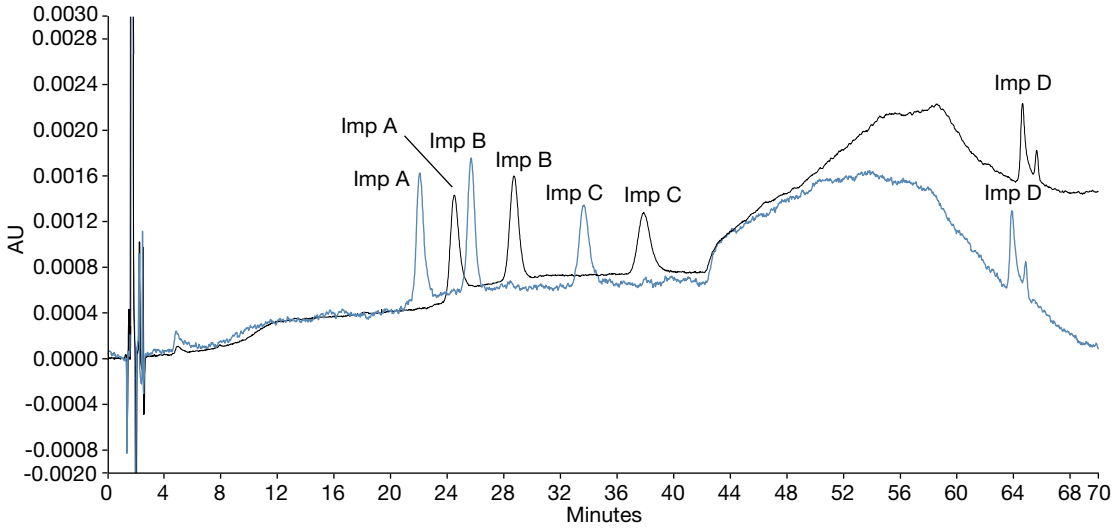

Figure 2. System suitability Atorvastatin impurity (imp) (Imp A, Imp B, Imp C, and Imp D) standard solutions on an Agilent 1260 system (blue) and a Vanquish Flex UHPLC system (black)

Table 3. System Suitability Target (SST) summary. Requirement: Resolution NLT (not less than) 1.5 between atorvastatin and related compound B from system suitability solution

		Agilent 1260 LC		Vanquish Flex UHPLC	
Solution	Compound	Resolution	Signal-to-noise ratio	Resolution	Signal-to-noise ratio
System suitability	Atorvastatin	1.8	-	1.7	-
Standard	RC A	-	33	-	64
	RC B	4.0	35	3.6	66
	RC C	7.7	20	6.4	38
	RC D	32.0	20	23.7	50

Conclusion

- The system suitability comparison of an Agilent 1260 Infinity LC system and a Thermo Scientific Vanquish Flex UHPLC system was demonstrated for the USP method for Atorvastatin Calcium impurity analysis.
- Although both systems met system suitability requirements, the Vanquish Flex UHPLC system showed improved sensitivity.

References

1. Mills, Simona, Analytical Method Transfer Checklist 2021, https://www.propharmagroup.com/blog/how-conduct-analytical-method-transfer/ (accessed January 25, 2022).
2. Thermo Scientific Case Study 000566: Method transfer onto the Thermo Scientific Vanquish HPLC/UHPLC platform: A CDMO perspective. https://assets.thermofisher.com/TFS-Assets/CMD/Reference-Materials/cs-000566-lc-method-transfer-vanquish-cdmo-cs000566-na-en.pdf
3. "The Top 300 of 2019", ClinCalc. https://clincalc.com/DrugStats/Top300Drugs.aspx (accessed October 16, 2021).
4. "Atorvastatin Calcium Monograph for Professionals", Drugs.com. AHFS. https://www.drugs.com/monograph/atorvastatin.html (accessed December, 23 2018).
5. World Health Organization. World Health Organization model list of essential medicines: 22nd list (2021). Geneva: World Health Organization. hdl:10665/345533. https://apps.who.int/iris/handle/10665/345533. WHO/MHP/HPS/EML/2021.02.
6. United States Pharmacopeia USP43-NF38, Atorvastatin Calcium Organic Impurities Procedure 1 method, The United States Pharmacopeial Convention, 2021.
7. United States Pharmacopeia USP43-NF38, General Chapter <621> Chromatography

Learn more at **thermofisher.com/vanquish**

thermo scientific