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Method Migration of the USP Ibuprofen Assay and Organic Impurities Method to an Alliance™ iS HPLC System

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Abstract

Method migration, or moving a method from one high-performance liquid chromatography (HPLC) system to another, is routine practice in many regulated laboratories, where there may be a variety of HPLC systems from different vendors and different models in a single lab. However, moving a method across systems can be challenging as differences between instrument designs may lead to chromatographic changes that can impact critical results. Alternatively, lack of familiarity with proper set up or operation of a system can also lead to poor results and may be much harder for the end user to identify. The Alliance iS HPLC System aims to make method migration more straightforward with improved usability features that limit analyst error and improve system reliability while maintaining chromatographic and quantitative performance. In this study a United States Pharmacopeia (USP) monograph method for the analysis of Ibuprofen and related impurities is replicated on two legacy HPLC systems and subsequently migrated to the Alliance iS HPLC System. Results are analyzed and compared, with all three systems demonstrating comparable results while meeting the USP system suitability requirements.

Benefits

- Ability to migrate USP monographs to the Alliance iS HPLC System, meeting system suitability requirements
- Consistent and reliable quantitative results
- Decreased user error

Introduction

Migration, or moving a method across two different LC systems, is an important consideration in any regulated laboratory. Modernization of analytical equipment is often necessary due to system availability and/or to take advantage of system improvements. Differences in systems can lead to changes in chromatography that may impact critical results. In addition, unfamiliarity with a system may lead to improper set up and/or operation of the system.

In this study an isocratic United States Pharmacopeia (USP) monograph for ibuprofen and related organic impurities is used to assess a method migration between HPLC systems. System suitability criteria, including peak area %RSD, retention time %RSD, USP tailing, signal-to-noise (s/n), and USP resolution will be evaluated. Analysis is conducted on two legacy HPLC systems as well as a modern HPLC, the Alliance iS HPLC System (Figure 1). These results demonstrate the ability to migrate methods to newer LC systems, enabling modernization of HPLC systems in the lab.

Figure 1. The Alliance iS HPLC System.

Experimental

All samples are prepared in accordance with USP 41-NF 36:

Assay

- Standard Solution consists of 10.0 mg/mL Ibuprofen (p/n: I4883-5G, Sigma-Aldrich) in mobile phase.

Organic Impurities

- Sensitivity Solution consists of 0.005 mg/mL Ibuprofen in mobile phase.
- Standard Solution consists of 0.2 mg/mL Ibuprofen, 0.01 mg/mL Ibuprofen Related Compound J (RCJ) (p/n: PHR1978-50MG, MilliporeSigma), and 0.01 mg/mL Ibuprofen Related Compound C (RCC) (p/n: PHR1146-500MG, MilliporeSigma) in mobile phase.
- System Suitability Solution consists of 10.0 mg/mL Ibuprofen, 0.01 mg/mL Ibuprofen Related Compound J (RCJ) (p/n: PHR1978-50MG, MilliporeSigma), and 0.01 mg/mL Ibuprofen Related Compound C (RCC) (p/n: PHR1146-500MG, MilliporeSigma) in mobile phase.

Sample

- Consists of a nominal concentration of 10.0 mg/mL Ibuprofen in mobile phase. Generic ibuprofen tablets purchased from a pharmacy and analyzed past expiry.

LC Conditions

LC system:	1. Legacy System 1 2. Legacy System 2 3. Alliance iS HPLC System
Detection:	1. 2489 UV/Vis Detector, 254 nm @ 2 points/second 2. Diode Array Detector (DAD), 254 nm @ 2.5 Hz 3. Dual Wavelength UV Detector, 254 nm @ 2 points/second
Vials:	TruView pH Control LCMS Certified Vials, p/n: 186005666CV
Column(s):	XBridge C_{18}, 250 x 4.6 mm, 5 µm (p/n: 186003117)

Column temperature:	25.0 °C
Sample temperature:	15.0 °C
Injection volume:	10.0 µL
Flow rate:	2.0 mL/min
Mobile phase:	4 g/L chloroacetic acid (p/n: C19627, MilliporeSigma) in 40:60 Milli-Q Water:Acetonitrile (ACN), pH 3.0
Method:	*Isocratic 10-minute method.*

Data Management

Chromatography Software:	Empower™ 3 FR 4

Results and Discussion

Method Migration from Legacy HPLC Systems of Assay and Organic Impurities

With the wide variety of HPLC systems available, laboratories often have systems from different manufacturers that were purchased over many years. With the introduction of modern HPLC systems into a laboratory, there is a need to ensure that validated methods will provide comparable results to those obtained on the legacy systems. In this study, the ibuprofen assay and the monograph for organic impurities are used to assess system performance on two legacy HPLC systems and an Alliance iS HPLC System.

For the analysis, each system was run using independent mobile phase and sample preparations. The

monographs were followed, and the same column used in all cases to reduce non-instrument related variability. Both the assay and the organic impurities method use the same mobile phase and column, with different samples specified for each.

Using the legacy methodology for calculating system suitability, which includes calculation of s/n from a segment of the baseline within the Sensitivity Solution injection, the results for both the assay and the organic impurities are presented in Tables 1 and 2. For the assay, which requires five replicate injections of the ibuprofen standard, the system suitability criteria were met for all systems, with all values well below specification (Table 1). The assay sample set was followed immediately on each system by the organic impurities sample set which includes (in order) injections of a sensitivity solution, a system suitability standard, and a standard. Figure 2 provides representative chromatograms of the Organic Impurities System Suitability Solution on each system. All three systems performed well, meeting all USP requirements for organic impurities (Table 2). The Alliance iS achieved the lowest peak area and retention time standard deviations for both standards and demonstrated superior resolution and s/n for the organic impurities standards. Repeatability of the Alliance iS HPLC System is demonstrated in Figure 3 using six replicate injections of Organic Impurities Standard Solution.

Table 1

USP Ibuprofen tablets assay				
Criteria	USP specification	System		
		Legacy system 1	Legacy system 2	Alliance iS HPLC System
Standard area % RSD	NMT 2.0%	0.1%	0.1%	0.1%
Standard retention time % RSD	NMT 2.0%	0.0%	0.1%	0.0%
Standard tailing factor	NMT 2.5	1.5%	1.7%	1.8%
% Ibuprofen in sample	90.0 - 110.0% of label claim	99.2%	104.6%	103.6%

Table 2

USP Ibuprofen tablets organic impurities system suitability				
Criteria	USP specification	System		
		Legacy system 1	Legacy system 2	Alliance iS HPLC System
Ibuprofen area RSD	NMT 6.0%	3.3%	0.6%	0.4%
RC J area RSD	NMT 6.0%	0.1%	0.3%	0.1%
RC C area RSD	NMT 6.0%	0.3%	0.1%	0.1%
Ibuprofen retention time RSD	NMT 6.0%	0.0%	0.1%	0.0%
RC J retention time RSD	NMT 6.0%	0.0%	0.1%	0.0%
RC C retention time RSD	NMT 6.0%	0.0%	0.1%	0.0%
System suitability solution				
Resolution (HH) RCJ - Ibuprofen	NLT 2.5	13.5	13.8	15.5
Resolution (HH) Ibuprofen - RC C	NLT 2.5	8.2	8.1	8.7
Sensitivity solution				
Signal/noise	NLT 10	12	52	79
Sample				
% RC J	NMT 0.2%	0.0%	0.0%	0.0%
% RC C	NMT 0.25%	0.00%	0.00%	0.00%
% Unspecified 1	NMT 0.2%	0.1%	0.1%	0.1%
% Unspecified 2	NMT 0.2%	0.7%	1.0%	1.1%
% Total Unspecified	NMT 1.5%	0.8%	1.1%	1.2%

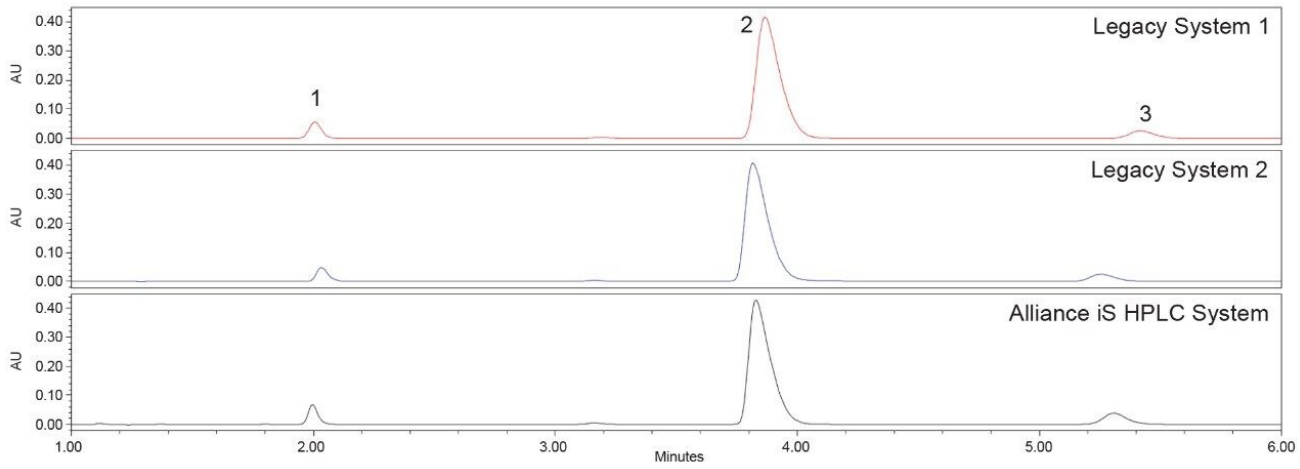

Figure 2. Representative chromatograms of the Organic Impurities System Suitability Solution on each system. Peak 1 = Related Compound J. Peak 2 = Ibuprofen. Peak 3 = Related Compound C.

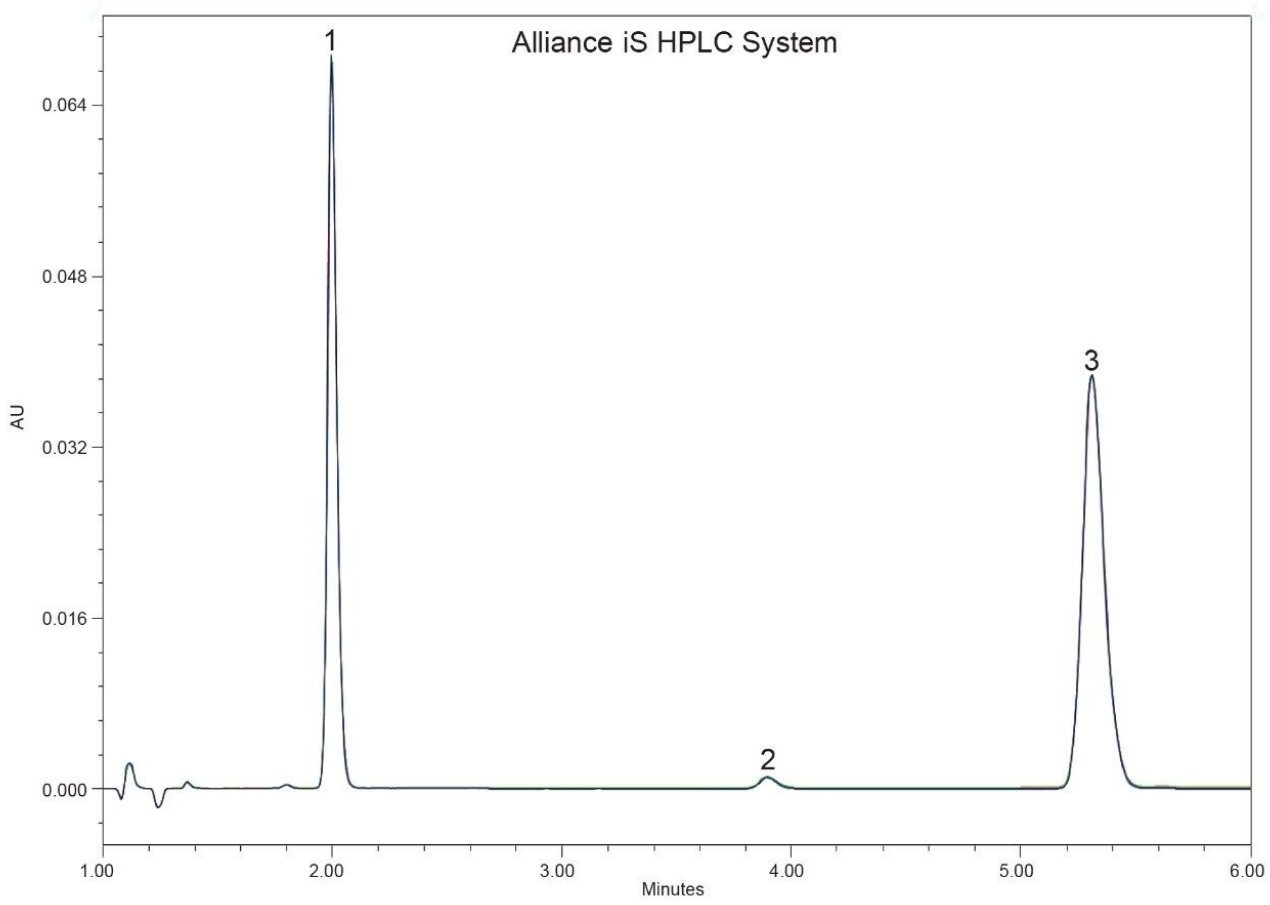

Figure 3. Overlaid chromatograms of the Organic Impurities Standard Solution on the Alliance iS HPLC System. N=6. Peak 1 = Related Compound J. Peak 2 = Ibuprofen. Peak 3 = Related Compound C.

Updating System Suitability Calculations for Resolution and Signal-to-Noise

Recent updates to 621 impact both USP resolution and USP s/n calculations.[1] USP resolution, which is reported in relation to baseline, was updated to be measured at half-height. Both values are reported by Empower with system suitability implemented. As shown in Table 2, the impact of reporting USP resolution at half-height does not impact the ability to meet system suitability criteria.

Along with updates to USP resolution, changes were made to determination of s/n. Historically, s/n could be calculated using either a blank injection or a stable baseline in the chromatogram.[2] For isocratic methods with ample stable baseline, using a stable section of the baseline in the chromatogram simplified processing. With

recent updates, it was recommended to use a blank injection and the noise at 20x or 5x the peak width at half height centered around the peak of interest (Figure 4). Given this guidance, the s/n was calculated for the organic impurities sensitivity sample on all three systems using all three methodologies (Table 3). With one legacy system and the Alliance iS HPLC Systems, the s/n criteria were met using all determination methodologies, with the lowest s/n observed using 20x peak width at half height. For the second legacy system, s/n at 20x peak width failed to meet system suitability of NLT 10 indicating challenges with making changes to historical USP calculations. It is important to note that the Alliance iS HPLC System demonstrated the highest s/n at the 20x measurement indicating superior baseline stability throughout the entire chromatogram compared to the legacy systems.

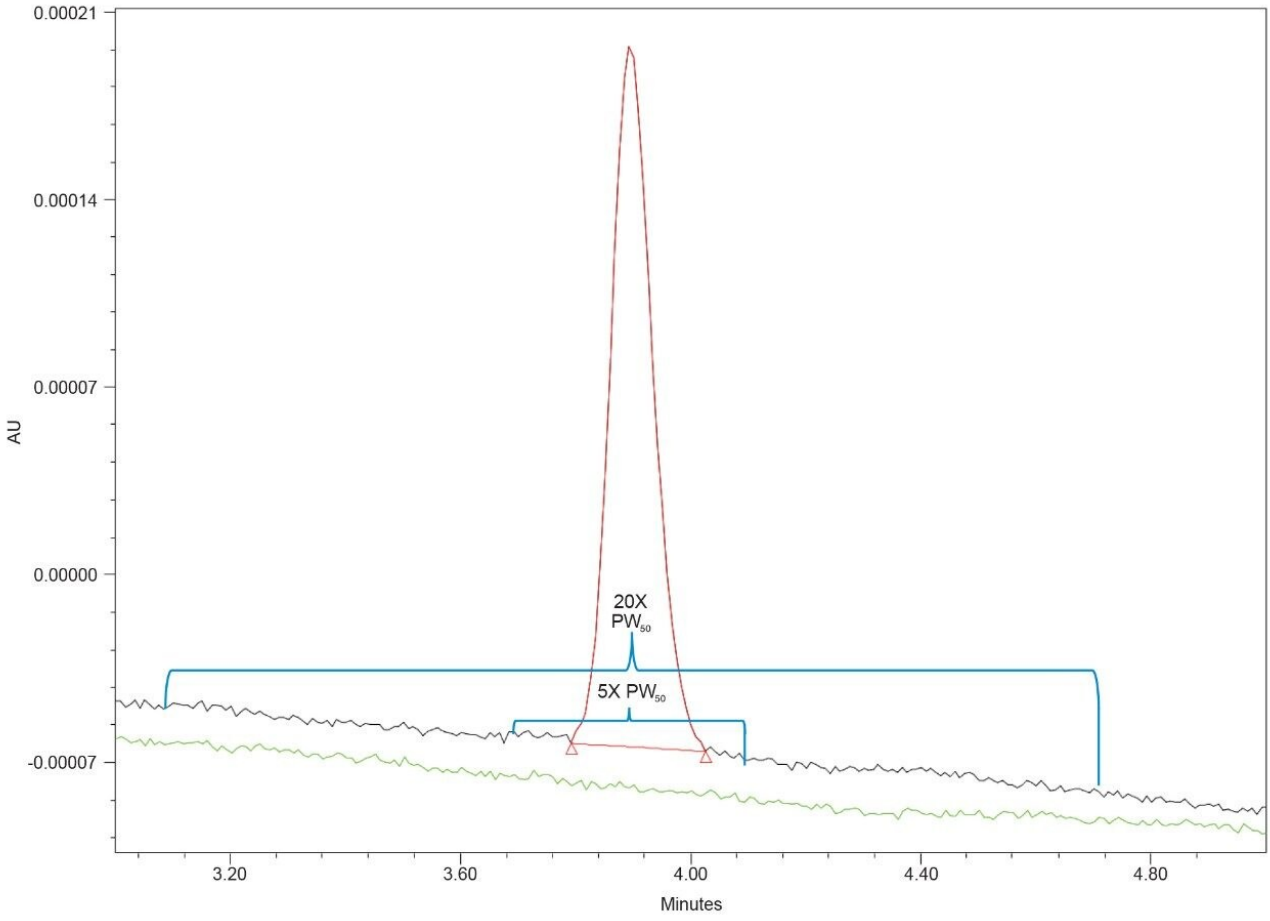

Figure 4. Overlaid chromatograms of the Sensitivity Solution and a blank injection on the Alliance iS HPLC System. PW_{50} = peak width at half height.

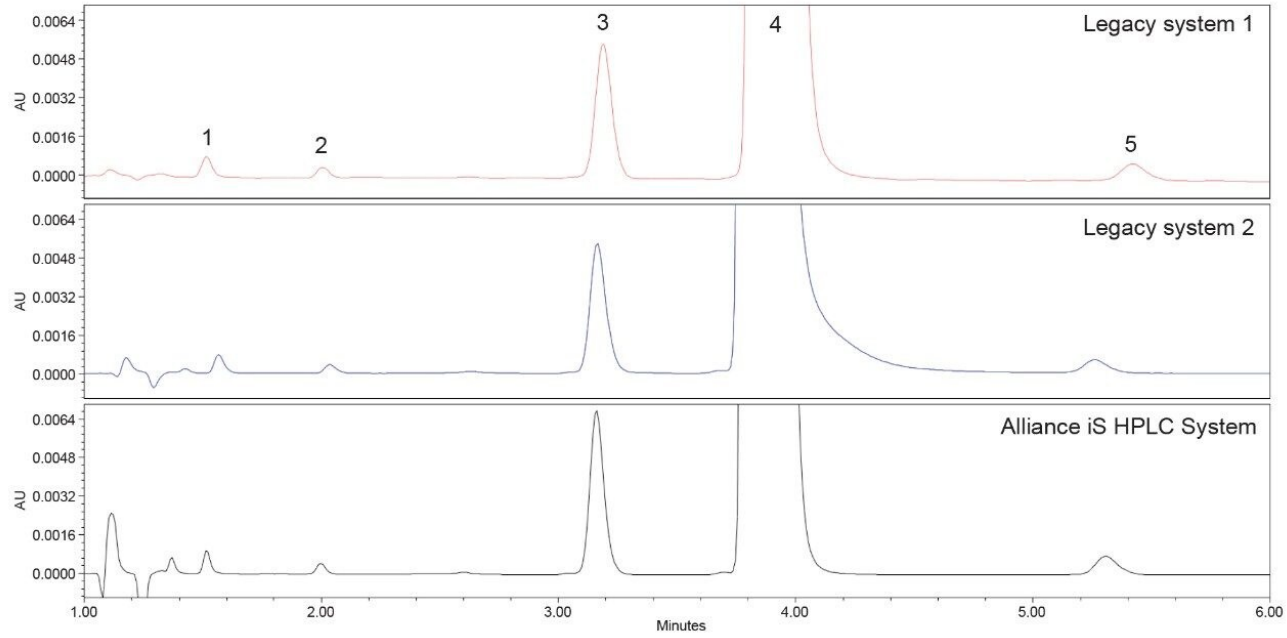

Figure 5. Stacked chromatograms of the Sample Solution run during the Organic Impurities portion of the analysis and used for known and unknown impurities measurements. Chromatograms are taken from the 2nd of 2 injections. Peak 1 = Unknown 1. Peak 2 = Related Compound J. Peak 3 = Unknown 2. Peak 4 = Ibuprofen. Peak 5 = Related Compound C.

Table 3

USP signal/noise				
Method	USP requirement	System		
		Legacy system 1	Legacy system 2	Alliance iS HPLC System
Within injection	NLT 10	12	52	79
5X PW_{50} in blank		21	94	84
20X PW_{50} in blank		7	28	39

Quantification of Organic Impurities in Sample

Quantification of a generic ibuprofen tablet sample was conducted on all three systems to demonstrate the

ability to migrate a method and achieve similar quantitative results. The USP monograph for Ibuprofen Tablets is designed to monitor the percentage of active ingredient in Ibuprofen tablets to ensure it is within the acceptable range of 90.0% and 110.0% of the labeled amount while also monitoring degradation products in solution.[3] Concentrations of degradation products must be monitored and reported, disregarding any peaks constituting less than 0.05% of the total peak area of the sample. When performing a method migration in a regulated environment, it is critically important that amounts of these degradation products remain consistent across systems as increases in reported amounts could lead to batch failures. Figure 5 displays representative chromatograms of Ibuprofen tablet samples run on each system and Table 2 reports the concentrations of each peak in solution. Concentrations of the three known peaks (Ibuprofen, Related Compound J, and Related Compound C) as well as the unknown peaks (unspecified degradation products) are consistent across systems.

Conclusion

Moving an established method across HPLC systems is often required due to system availability or modernization of a laboratory. In this work, a well-tested USP method for the analysis of Ibuprofen and organic impurities was analyzed on two legacy HPLC systems and the Alliance iS HPLC System. The Alliance iS HPLC System met all system suitability criteria, including signal-to-noise and provided comparable quantitative results to both legacy systems for both assay purity and quantitation of impurities in a generic formulation of Ibuprofen tablets. These results indicate the Alliance iS HPLC System allows for easy method migration from legacy instrumentation while achieving high performance and laboratory modernization.

References

1. GUID-6C3DF8B8-D12E-4253-A0E7-6855670CDB7B_6_en-US; United States Pharmacopeia, 2022.
2. GUID-6C3DF8B8-D12E-4253-A0E7-6855670CDB7B_1_en-US; United States Pharmacopeia, 2017.
3. GUID-B108D32E-F28B-41A3-84C7-62D1C4F37646_1_en-US; United States Pharmacopeia, 2016.

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