

Demonstration of the USP Quetiapine Fumarate Impurities Method Across a Wide Range of Liquid Chromatographic Systems

Amanda B. Dlugasch, Jennifer Simeone, and Patricia R. McConville
 Waters Corporation, Milford, MA, USA

APPLICATION BENEFITS

- The USP quetiapine fumarate impurity method can be effectively analyzed on the Alliance™ HPLC, the ACQUITY™ Arc™ UHPLC, and the ACQUITY UPLC™ H-Class PLUS Systems.
- Gradient elution methods can be run on a variety of liquid chromatography systems.
- The impurity results of a sample can be replicated on the Alliance HPLC System, the ACQUITY Arc UHPLC System, and the ACQUITY UPLC H-Class PLUS System.

WATERS SOLUTIONS

[Alliance HPLC System](#)

[ACQUITY Arc System](#)

[ACQUITY UPLC H-Class PLUS System](#)

[XBridge™ Columns](#)

[Empower™ 3 Chromatography Data Software](#)

KEYWORDS

ACQUITY UPLC H-Class PLUS, ACQUITY Arc UHPLC, Alliance HPLC, USP Methods, quetiapine fumarate, impurities, gradient elution

INTRODUCTION

Many pharmaceutical companies adhere to the United States Pharmacopeia (USP) methods when analyzing impurities contained within their active pharmaceutical ingredient (API). Many of these methods use gradient elution for the separation of impurity compounds. When a USP method contains a gradient elution it is important to consider which type of liquid chromatography system to use for the analysis. Based upon method conditions that consist of larger column dimensions and long run times, it may seem that the method needs to be run on a high performance liquid chromatography (HPLC) system, when in reality the method can be effectively run on a variety of systems. As long as the system suitability requirements of the USP method are met, the analysis can be completed on a liquid chromatography system that has lower dispersion and higher pressure specifications than an HPLC system, for example, UHPLC and UPLC systems.

In this application the USP quetiapine fumarate impurity (Figure 1) method¹ will be analyzed on three different types of liquid chromatography systems, including the Alliance HPLC System, the ACQUITY Arc UHPLC System, and the ACQUITY UPLC H-Class PLUS System. The successful execution of the method on each system will be evaluated based upon the system suitability requirements listed in the USP monograph.

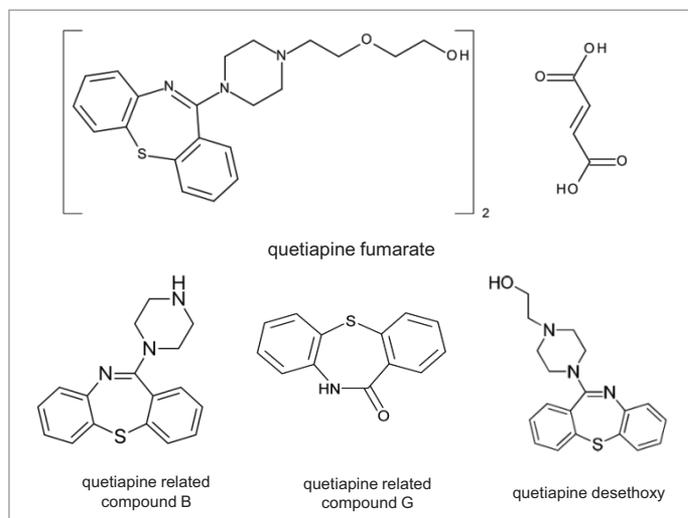


Figure 1. Structure of quetiapine fumarate (API) and its impurities of quetiapine related compound B, quetiapine related compound G, and quetiapine desethoxy.

EXPERIMENTAL

Sample description

The Quetiapine Fumarate Standard (Catalog#: 1592704), the Quetiapine System Suitability Standard (Catalog#: 1592715), Quetiapine Related Compound B (Catalog#: 1592737) and Quetiapine Related Compound G (Catalog#: 1592781) were purchased from the United States Pharmacopeia. The unknown quetiapine fumarate sample was purchased from Alibaba.com.

All solutions were prepared to the designated concentrations per the USP monograph. The system suitability, standard, and peak identification solutions were prepared in the diluents of solution A: solution B (86:14), where solution A is acetonitrile and buffer (25:75) and solution B is acetonitrile. The sample solution was prepared in solution A.

The concentrations of the solutions are 1.0 mg/mL for the system suitability solution, 0.001 mg/mL for the standard solution, and 1.0 mg/mL for the sample solution. The peak identification solution is 1 µg/mL of USP Quetiapine Fumarate Standard, 10 µg/mL of USP Quetiapine Related Compound B, and 2 µg/mL of USP Quetiapine Related Compound G.

Method conditions

LC conditions

LC systems: Alliance e2695 Separations Module with 100 µL syringe, 2998 PDA Detector and CH-30 equipped with passive column preheater
ACQUITY Arc System with active solvent preheating (CH-30A) and 2998 PDA Detector (Path 1)
ACQUITY UPLC H-Class PLUS System with active solvent preheating (CH-30A), 50 µL extension loop and ACQUITY UPLC PDA Detector

Column: XBridge BEH C8 Column, 3.5 µm, 4.6 mm × 150 mm (p/n: [186003055](#))

Column temp.: 45 °C

Sample temp.: 4 °C

Injection volume: 20 µL

Flow rate: 1.5 mL/min

Run time: 70 minutes

PDA wavelength: 250 nm at 4.8 nm resolution

Data rate: 2 points/sec (Hz)

Mobile phase A: Acetonitrile and buffer (25:75)

Mobile phase B: Acetonitrile

Buffer: 3.1 g/L of ammonium acetate in water. 2 mL of 25% ammonium hydroxide was added to each 1 L of solution. The final pH is not less than (NLT) 9.2

Time (min)	Mobile phase A (%)	Mobile phase B (%)
0.0	100.0	0.0
25.0	100.0	0.0
60.0	29.3	70.7
60.1	100.0	0.0
68.0	100.0	0.0

Table 1. Gradient.

Data management

Empower 3 Chromatography Data Software, FR 3

RESULTS AND DISCUSSION

The USP method for the impurities of quetiapine fumarate was analyzed on the Alliance HPLC, the ACQUITY Arc UHPLC, and the ACQUITY UPLC H-Class PLUS systems using the conditions specified in the monograph. The peak identification solution, the system suitability solution, the standard solution and an unknown sample solution were prepared as described in the monograph and were analyzed in six replicates. Each system was evaluated based upon the system suitability requirements and the acceptance criteria for the sample impurities outlined in the monograph.

The peak identification solution provides proper identification of the three components, quetiapine, quetiapine related compound B, and quetiapine related compound G, contained in the solution. Identification was based on information provided in the monograph including relative retention times and peak size. An advantage of running the impurity method on the lower dispersion systems is the decrease in peak width resulting in an increase in the peak height,² which may be able to provide lower detection limits of the smaller impurity peaks. The system dispersion values determined for the systems are 44.5 μ Ls for the Alliance HPLC System, 21.5 μ Ls for the ACQUITY Arc UHPLC System, and 9.6 μ Ls for the ACQUITY UPLC H-Class PLUS System. The increase in peak height and decrease in peak width can be seen with the Peak ID solution (Figure 2). All three impurity compounds increase in peak height and decrease peak width as the dispersion of the LC system is decreased from the Alliance HPLC system to the ACQUITY UPLC H-Class PLUS System (Figure 2).

The resolution between two closely eluting peaks demonstrates the efficiency of a method on a system.³ For the USP impurity method, the system suitability solution is used to evaluate the resolution between two different critical pairs. The monograph requires that the resolution between the quetiapine desethoxy and quetiapine peaks be not less than (NLT) 4.0 and the resolution is NLT 3.0 between the quetiapine related compound B and quetiapine related compound G peaks (Figure 3). The results of the system suitability solution on all three systems meet the USP requirements and can be found in Table 2.

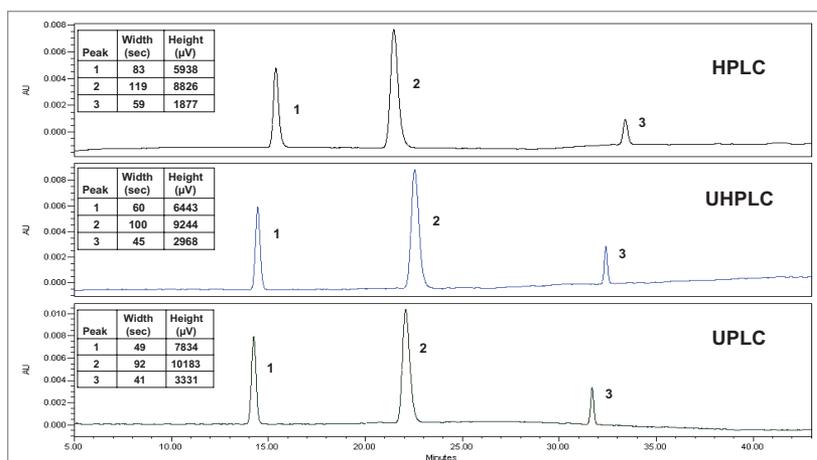


Figure 2. Peak ID solution chromatograms, with the corresponding peak width and peak heights, for the three impurity compounds obtained using each LC system. The compounds were identified based on the relative retention time provided in the USP method. The compounds are identified as follows: 1: quetiapine related compound G 2: quetiapine related compound B, 3: quetiapine.

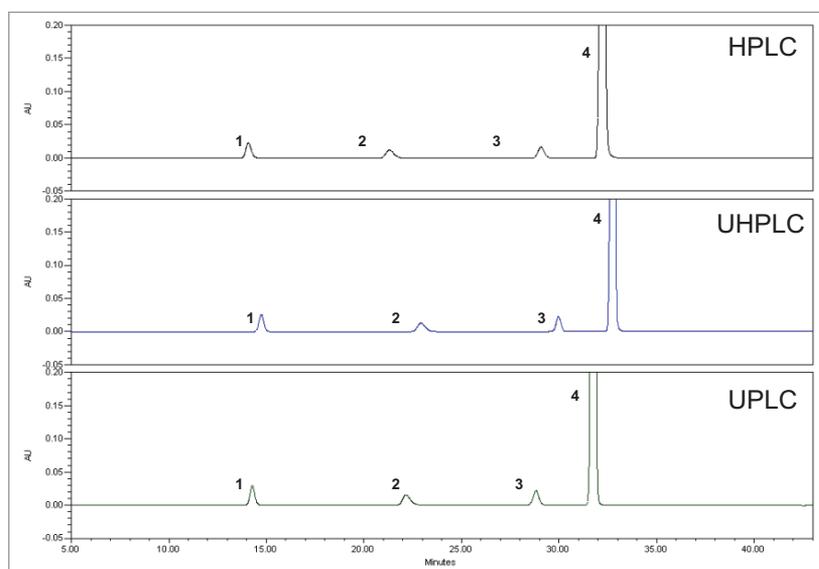


Figure 3. System suitability solution run on three LC instruments. The compounds were identified based on the relative retention time provided in the USP method. The compounds are identified as follows: 1: quetiapine related compound G, 2: quetiapine related compound B, 3: quetiapine desethoxy, and 4: quetiapine.

Table 2. Resolution results for the system suitability solution on all three LC systems.

	Resolution between quetiapine desethoxy and quetiapine peaks	Resolution between compound B and compound G peaks
Alliance HPLC System	6.6	11.0
ACQUITY Arc UHPLC System	7.4	13.3
ACQUITY UPLC H-Class PLUS System	6.9	13.4
USP requirements	NLT 4.0	NLT 3.0

The reproducibility of a system and peak symmetry are evaluated by examining the percent relative standard deviation (%RSD) and tailing factor of six replicate injections.⁴ The monograph requires a tailing factor of not more than (NMT) 2.0 and %RSDs for area and retention time be NMT 5.0% for the quetiapine peak contained in the standard solution (Figure 4). The results from each system for the standard solution can be found in Table 3, and all systems successfully met the tailing factor and %RSD criteria.

The analysis of the unknown sample requires injection of the standard solution at a known concentration (Figure 4). The percent impurity contained within the unknown sample is determined using the following equation:

$$\text{Result} = (r_u/r_s) \times (C_s/C_u) \times (1/F) \times 100$$

Where r_u is the peak response of each impurity for the sample solution, r_s is the peak response of quetiapine from the standard solution, C_s is the concentration of USP quetiapine fumarate standard in the standard solution (mg/mL), C_u is the concentration of quetiapine fumarate in the sample (mg/mL), and F is the relative response factor provided in the monograph.¹ Analysis of the unknown sample yielded two impurity peaks. One was identified as quetiapine desethoxy, while the other peak could not be identified. The % impurity was calculated using the response factor provided in the monograph where applicable. The calculated amount of the quetiapine desethoxy impurity and the unknown impurity are both below the acceptance criteria value set in the monograph. Additionally, the calculated concentration of the quetiapine desethoxy impurity and the unknown impurity are consistent and reproducible across all three chromatographic systems (Table 4).

Table 3. Tailing factor and RSD results for the standard solution on the three systems.

	Tailing factor	Retention time %RSD	Area %RSD
Alliance HPLC System	1.0	0.14	0.84
ACQUITY Arc UHPLC System	1.0	0.05	1.14
ACQUITY UPLC H-Class PLUS System	1.0	0.10	1.18
USP requirements	NMT 2.0	NMT 5.0%	NMT 5.0%

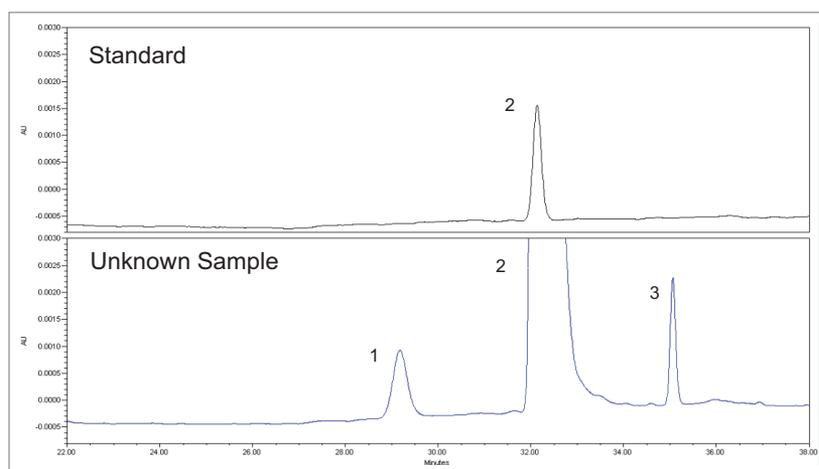


Figure 4. Standard solution (0.001 mg/mL) containing quetiapine (top) and the unknown sample solution (1.0 mg/mL) (bottom) run on the Alliance HPLC System. The compounds were identified based on the relative retention time provided in the USP method. The compounds are identified as follows: 1: quetiapine desethoxy 2: quetiapine and 3: unknown impurity.

Table 4. Calculated percent of the impurity of quetiapine desethoxy and the unknown impurity peak in the unknown sample on the three LC systems.

	Quetiapine desethoxy	Unknown impurity
Alliance HPLC System	0.10%	0.07%
ACQUITY Arc UHPLC System	0.13%	0.08%
ACQUITY UPLC H-Class PLUS System	0.12%	0.08%
USP acceptance criteria	NMT 0.15%	NMT 0.10%

CONCLUSIONS

Many USP gradient elution methods use columns with packing material of >3 μm particle size that can be effectively analyzed on a variety of LC systems. The USP quetiapine fumarate impurity method was successfully analyzed on three different LC systems: an Alliance HPLC System, an ACQUITY Arc UHPLC System and an ACQUITY UPLC H-Class PLUS System. The analysis on all three LC systems met the system suitability requirements and provided consistent and reproducible impurity concentration values for an unknown sample.

References

1. Official Monographs, Quetiapine Fumarate, USP 40 NF35 S1, United States Pharmacopeia and National Formulary (USP 40-NF35 S1) Baltimore, MD: United Book Press, Inc.; 2017. p. 5939.
2. Hong, Paula; McConville, Patricia. Dwell Volume and Extra-Column Volume: What Are They and How Do They Impact Method Transfer. Waters White Paper [720005723EN](#). 2016 Oct.
3. Snyder, Lloyd R.; Kirkland, Joseph; Dolan, John W. *Introduction to Liquid Chromatography*, 3rd Edition. Hoboken: Wiley and Sons, 2010. Section 2.5 p54–57. Print.
4. Chapter <621> *Chromatography*. United States Pharmacopeia and National Formulary (USP 40-NF35 S1) Baltimore, MD: United Book Press, Inc.; 2017.

Waters

THE SCIENCE OF WHAT'S POSSIBLE.™

Waters, The Science of What's Possible, ACQUITY, UPLC, Alliance, Arc, Empower, and XBridge are trademarks of Waters Corporation. All other trademarks are the property of their respective owners.

©2019 Waters Corporation. Produced in the U.S.A. February 2019 720006292EN AG-PDF

Waters Corporation
34 Maple Street
Milford, MA 01757 U.S.A.
T: 1 508 478 2000
F: 1 508 872 1990
www.waters.com