APPLICATIONS

Related Substances Assay of the Drug Product Clarithromycin on a Kinetex 3.5 µm XB-C18 Column According to Ph. Eur. Monograph 1651

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In addition to chromatography, Dr. Behr has an absolute love for heavy metal music and skiing in the Alps.

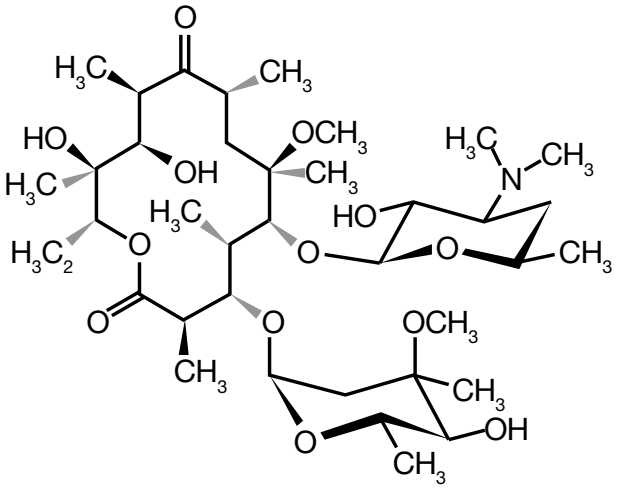
In this technical note, we report the separation between clarithromycin and its related impurities using a Kinetex® 3.5µm XB-C18 HPLC column according to the Ph. Eur. Monograph 1651 related substances assay. Comparison with an AkzoNobel® Kromasil® 3.5µm C18 is also provided.

Introduction

The European Pharmacopoeia (Pharmacopoea Europaea, Ph. Eur.) is a single reference work for the quality control of medicines in the signatory states of the Convention on its elaboration. The official standards published within provide a legal and scientific basis for quality control during the development, production, and marketing processes. They concern the qualitative and quantitative composition and the tests to be carried out on medicines, on the raw materials used in production of medicines and on the intermediates of synthesis. All producers of medicines and/or substances for pharmaceutical use must therefore apply these quality standards in order to market their products.

Clarithromycin is an antibiotic used to treat a variety of bacterial infections including strep throat, pneumonia, skin infections and Lyme disease.

Figure 1.
Clarithromycin



Materials and Methods

The analysis was performed using an Agilent® 1100 equipped with a multiple wavelength length UV detector. The Kinetex 3.5µm XB-C18 column used for analysis was obtained from Phenomenex (Torrance, CA, USA). The Kromasil 3.5µm C18 used for analysis was obtained from AkzoNobel, Separation Products (SE-445 80 Bohus, Sweden). All solvents were purchased from Honeywell (Morristown, NJ, USA) and Sigma-Aldrich (St. Louis, MO, USA). Ph. Eur. Standard Clarithromycin for peak identification CRS Y0000321 was purchased from European Directorate for the Quality of Medicines & HealthCare (EDQM) – Council of Europe; Postal address: 7 Allée Kastner CS 30026 F - 67081 STRASBOURG (France).

Ph. Eur. Monograph 1651

Ph. Eur. Monograph 1651 system suitability reference solution D. Dissolve 15 mg of Clarithromycin for peak identification CRS in 5 mL of acetonitrile and dilute to 10 mL with water.

Column:
- *size: l = 0.10 m, Ø = 4.6 mm*
- *stationary phase: octadecylsilyl silica gel for chromatography R (3.5 µm)*
- *temperature: 40 °C*

Mobile phase:
- *A: a 4.76 g/L solution of potassium dihydrogen phosphate adjusted to pH 4.4 with dilute phosphoric acid*
- *B: Acetonitrile*

Gradient: 25 % mobile phase B to 60 % mobile phase B in 32 min, hold for 2 min

Flow rate: 1.1 mL/min

Detection: spectrophotometer at 205 nm

Injection: 10 µL

Relative retention with reference to Clarithromycin (retention time = about 11 min): impurity A = about 0.42; impurity J = about 0.63; impurity L = about 0.74; impurity B = about 0.79; impurity M = about 0.81; impurity C = about 0.89; impurity D = about 0.96; impurity N = about 1.15; impurity E = about 1.27; impurity F = about 1.33; impurity P = about 1.35; impurity O = about 1.41; impurity K = about 1.59; impurity G= about 1.72; impurity H = about 1.82.

System suitability: reference solution D:
- *peak-to-valley ratio: minimum 3.0, where Hp= height above the baseline of the peak due to impurity D and Hv = height above the baseline of the lowest point of the curve separating this peak from the peak due to clarithromycin in the chromatogram obtained with reference solution D.*

Result and Discussion

In this tech note we report the separation between Clarithromycin (the active pharmaceutical ingredient) and its related impurities per the related substances section of Ph. Eur. Monograph 1651, as shown in **Figure 2**. On the Kinetex column the Clarithromycin peak elutes at 9.25 min. The peak-to-valley ratio between impurity D and clarithromycin is 10.8 which passes the system suitability minimum requirement of 3.0. As a comparison, a Kromasil® 3.5 µm C18 was also used for the analysis, as shown in **Figure 3**. On the Kromasil column the Clarithromycin peak elutes later at 11.2 minutes. The peak-to-valley ratio between impurity D and clarithromycin on the Kromasil column is 10.0, slightly less than on the Kinetex column.

Conclusion

The results shown above demonstrate that a Kinetex 3.5 µm XB-C18, 100 x 4.6 column can be successfully used to analyze Clarithromycin according to Ph. Eur. Monograph 1651. The peak-to-valley ratio between the API and impurity D for system suitability was 10.8 which is well above the minimum of 3.0 required by the Ph. Eur. monograph method. In comparison to the Kromasil 3.5 µm C18, the Kinetex column acquired a faster analysis with slightly improved resolution between clarithromycin and the closely eluting impurity D.

Figure 2.
Clarithromycin reference solution D on a Kinetex 3.5 µm XB-C18, 100 x 4.6 (Peak-to-valley ratio is 10.8)

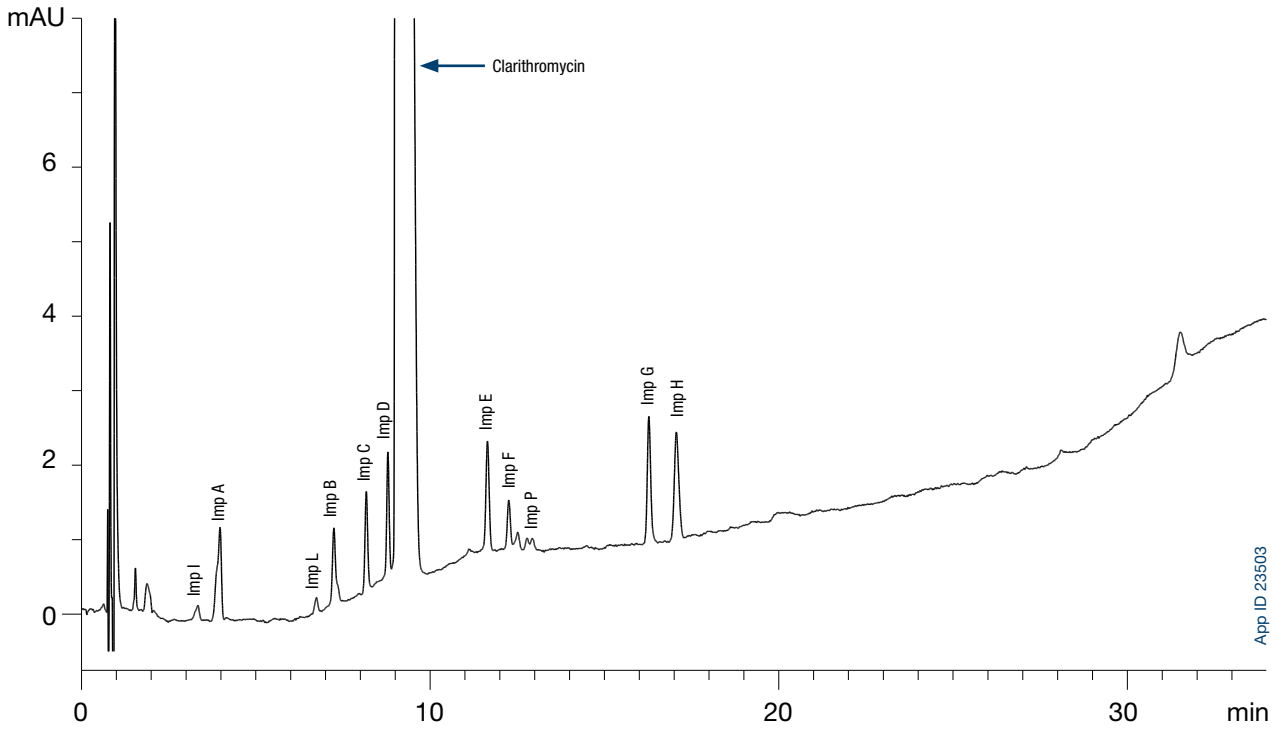
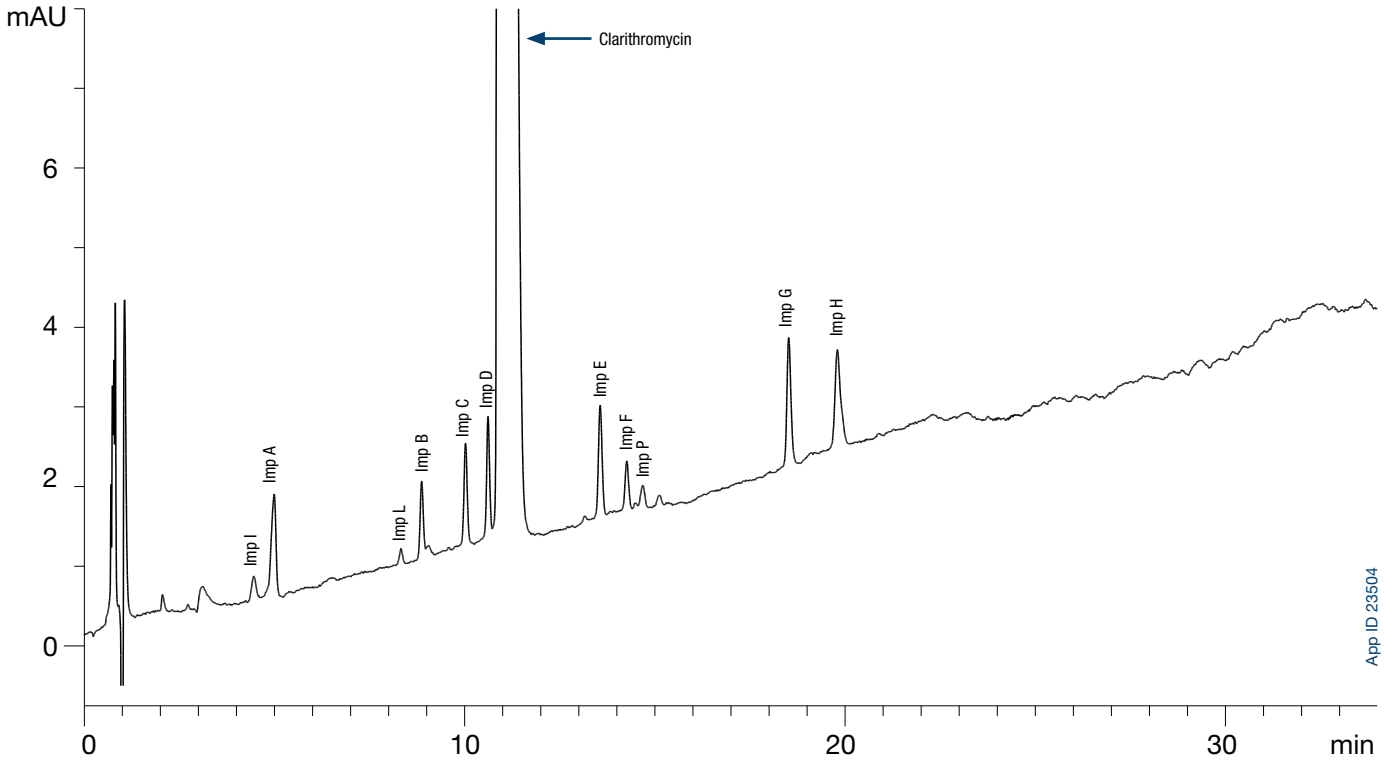


Figure 3.
Clarithromycin reference solution D on a Kromasil 3.5 µm C18, 100 x 4.6 (Peak-to-valley ratio is 10.0)



Kinetex 3.5 µm Analytical Columns	SecurityGuard ULTRA Cartridges (mm)
Phases	100 x 4.6 mm	3pk
XB-C18	00F-4744-E0	AJ0-8768 for 4.6 mm ID

* SecurityGuard ULTRA Cartridges require holder, Part No.: AJ0-9000

guarantee

If Phenomenex products in this technical note do not provide at least an equivalent separation as compared to other products of the same phase and dimensions, return the product with comparative data within 45 days for a FULL REFUND.

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