# APPLICATION BRIEF



# Liquid Chromatography

# HPLC Analysis of the Semi-Synthetic Penicillin, Amoxicillin

# Introduction

Antibiotics are natural substances released by bacteria and fungi which are capable of killing, or inhibiting, competing microbial species. This phenomenon has long been known; but it was not until 1928 that penicillin, the first true antibiotic,

was discovered by Alexander Fleming, Professor of Bacteriology at St. Mary's Hospital in London.<sup>1</sup> However, it 12 years later that pen years later before penicillin was isolated and developed as a medicine by Howard Florey and Ernst Chain.<sup>2</sup>

Since this breakthrough in therapeutic medicine, derivates of penicillin were developed with increased efficacy levels. Amoxicillin is one of several such semisynthetic penicillin's. First discovered in the 1960's its patent has now expired. Consequently, amoxicillin and co-amoxiclav is now marketed under many different trade names worldwide. It is commonly used in the treatment of numerous infections including pneumonia, skin infections and Lyme disease.

This application brief illustrates the rapid analysis of amoxicillin, Figure 1, using the Quasar C18 column.



# **Experimental Conditions**

### **Method Parameters**

All LC method parameters are shown in Table 1.

#### Table 1. LC Method Parameters.

Quasar C18	100 mm	4.6 mm	3 µm	N9308806		
Mobile Phase	A: H <sub>2</sub> 0 (+0.1% formic acid) B: ACN (+0.1% formic acid) Gradient: 5-50% B in 10 minutes					
Flow Rate	1 mL/min					
Тетр	20 °C					
Wavelength	254 nm					
Injection Vol.	5 μΙ					
Analyte	Amoxicillin					

# Column Used:

# Solvents and Samples

All solvents were HPLC grade and samples were filtered using a  $0.45\mu m$  nylon filter.

# **Results and Discussion**

Amoxicillin, Figure 1, is successfully analysed in just over 5 minutes using the Quasar C18 column, 100 mm in length, Figure 2. Ideally suited to the analysis of small molecules, such as this antibiotic, the Quasar C18 phase provides enough retention via ligand/analyte interactions whilst also maintaining peak shape due to the ultra-high purity silica base and low residual silanol activity.



Figure 1. Chemical Structure of Amoxicillin.





# Conclusion

- The Quasar C18 HPLC phase offers high efficiency separation of this semi-synthetic antibiotic.
- The ultra-high purity silica base and low residual silanol activity yields excellent peak shape even for basic analytes, such as amoxicillin.
- Run time could be further reduced by switching to UHPLC and using a Quasar C18 1.7 µm column.

### References

- Haven KF (1994). Marvels of Science : 50 Fascinating 5-Minute Reads. Littleton, CO: Libraries Unlimited. p. 182. ISBN 978-1-56308-159-0.
- "Making Penicillin Possible: Norman Heatley Remembers". ScienceWatch. Thomson Scientific. 2007. Archived from the original on February 21, 2007. Retrieved 2007-02-13.

# **Consumables**

		Part N	Part Number		
Nylon filters	02542	02542880			
Phase	Length (mm)	I.D. (mm)	μm	Part	
Quasar C18	300	3.9	5	N9308800	
Quasar C18	250	4.6	5	N9308801	
Quasar C18	150	4.6	5	N9308802	
Quasar C18	100	4.6	5	N9308803	
Quasar C18	50	4.6	5	N9308804	
Quasar C18	150	4.6	3	N9308805	
Quasar C18	100	4.6	3	N9308806	
Quasar C18	50	4.6	3	N9308807	
Quasar C18	150	3.0	3	N9308808	
Quasar C18	100	3.0	3	N9308809	
Quasar C18	50	3.0	3	N9308810	
Quasar C18	150	2.1	3	N9308811	
Quasar C18	100	2.1	3	N9308812	
Quasar C18	50	2.1	3	N9308813	
Quasar C18	100	4.6	1.7	N9308814	
Quasar C18	50	4.6	1.7	N9308815	
Quasar C18	100	3.0	1.7	N9308816	
Quasar C18	50	3.0	1.7	N9308817	
Quasar C18	100	2.1	1.7	N9308818	
Quasar C18	50	2.1	1.7	N9308819	

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