

Analysis of Lamivudine Using a Core Enhanced Technology Accucore HPLC Column

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Key Words

- Accucore aQ
- Fused core
- Superficially porous
- Lamivudine
- USP
- FDA

Abstract

This application note demonstrates the use of Thermo Scientific Accucore aQ, for the fast HPLC analysis of lamivudine. The method of analysis was adapted from the USP monograph, by using an in-house method transfer calculator [1].

Introduction

Accucore™ HPLC columns use Core Enhanced Technology to facilitate fast and high efficiency separations. The 2.6 µm diameter particles consist of a solid core and a porous outer layer. The optimised phase bonding creates a series of high coverage, robust phases. Accucore aQ is a C18 phase with polar endcapping, which provides a controlled interaction mechanism by which polar analytes can be retained; the endcapping is responsible for the phase stability in highly aqueous mobile phases.

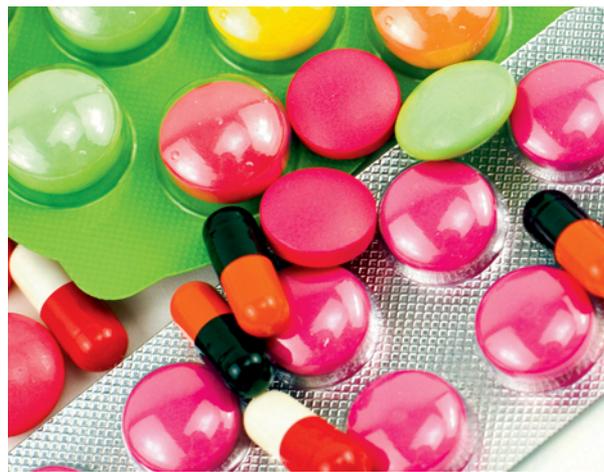
The tightly controlled 2.6 µm diameter of Accucore particles results in much lower backpressures than typically seen with sub-2 µm materials.

Lamivudine (tradename: Zeffix, Heptovir, Epivir), is an antiretroviral drug used for the treatment of hepatitis B and HIV. It acts by stopping the viral DNA growth, but it can not help cells already infected. It is an FDA approved medication, often given in combination with zidovudine. The United States Pharmacopeia (USP) provides worldwide guidance for the chromatographic analysis of lamivudine [2], which is based on High Performance Liquid Chromatography (HPLC). The implementation of Accucore aQ, allowed for the lamivudine to be analyzed according to the USP monograph.

Experimental details

The analysis was run on a Thermo Scientific Accela UHPLC system. The data were acquired and processed using Thermo Scientific ChromQuest 5.0 Software.

The separation of lamivudine was achieved on a Core Enhanced Technology Accucore aQ HPLC column.



Sample Preparation

A 700 µg/mL of lamivudine standard solution was prepared in 50:50 methanol: water; this solution was then diluted to 35 µg/mL in water and used for the analysis.

Thermo Scientific Column	Part Number
Accucore aQ 2.6 µm 100 x 2.1 mm	17326-10213
Measured Pressure 109 bar	

Thermo Scientific HPLC system

Column temperature	35 °C
Injection volume	1.0 µL
Flow rate	0.2 mL/min
UV detection	277 nm

Mobile Phase

95:5 ammonium acetate (25 mM, pH= 3.8):methanol

Consumables	Part Number
Fisher Scientific HPLC grade water	W/0106/17
Fisher Scientific HPLC grade methanol	M/4056/17
Fisher Scientific Ammonium acetate	A/3440/50
Thermo Scientific 2 mL clear vial and Si/PTFE seal	60180-600

Results

The original USP analytical conditions, based on a 250 x 4.0 mm, 5 µm column were scaled down using our method transfer calculator to accommodate for the column geometry reduction. The analysis was carried out on an Accucore aQ 2.6 µm 100 x 2.1 mm column. As shown on Figure 1, lamivudine eluted at 2.4 min. The USP acceptance criteria (Tailing factor, $T_f \leq 2.0$, % RSD (for t_r and Peak Area) ≤ 2.0) were met, as demonstrated in Table 1. The statistical assessment is based on data from 6 replicate injections (see Table 1 for mean values).

Conclusions

The use of an Accucore aQ HPLC column, allowed to successfully scale down the USP method for the analysis of lamivudine, in order to increase sample throughput. The analytical results exceeded the specifications stated in the

USP monograph. Accucore aQ columns are therefore an excellent choice for the fast analysis of lamivudine, allowing a high sample throughput.

References

- [1] <http://www.hplctransfer.com/>
- [2] http://www.pharmacopeia.cn/v29240/usp29nf24s0_m4423.html

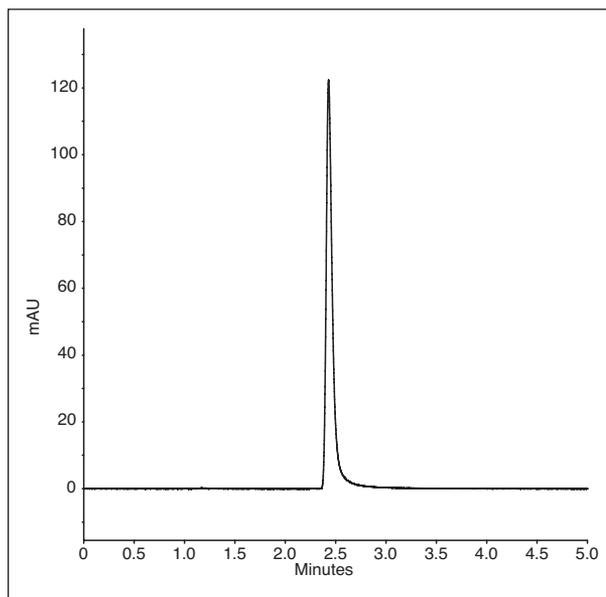


Figure 1: Chromatogram of 35 µg/mL of lamivudine separated on an Accucore aQ 2.6 µm 100 x 2.1 mm column

	t_r (min)	Peak Area	T_f
Mean	2.43	518396	1.58
%RSD	0.03	0.95	1.08

Table 1: Method Precision (%RSD) of chromatographic parameters for the analysis of lamivudine on an Accucore aQ 2.6 µm 100 x 2.1 mm column (data calculated from six replicate injections)

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