

Analysis of Zidovudine Using a Core Enhanced Technology Accucore HPLC Column

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Key Words

- Zidovudine
- USP
- HIV
- Accucore C18
- Fused core
- Superficially porous
- Core Enhanced Technology

Abstract

This application note demonstrates the use of Thermo Scientific Accucore C18, for the fast HPLC analysis of Zidovudine, allowing an increase in sample throughput. The method of analysis was adapted from the USP monograph, by using an in-house method transfer calculator [1].

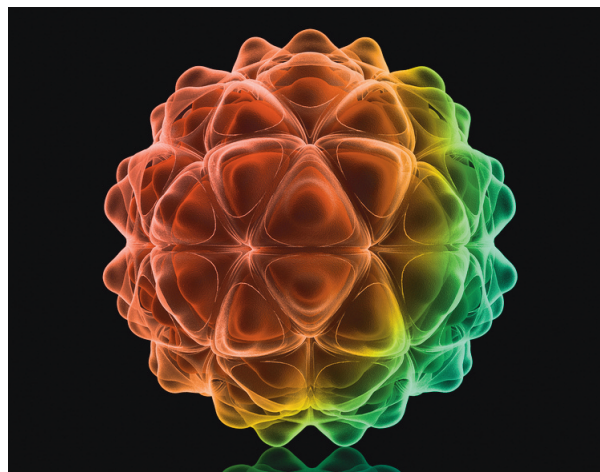
Introduction

Accucore™ HPLC columns use Core Enhanced Technology to facilitate fast and high efficiency separations. The 2.6 µm diameter particles are not totally porous, but rather have a solid core and a porous outer layer. The optimised phase bonding creates a series of high coverage, robust phases. The carbon loading of Accucore C18 provides high retention of non-polar analytes via a predominantly hydrophobic interaction mechanism. The tightly controlled 2.6 µm diameter of Accucore particles results in much lower backpressures than typically seen with sub-2 µm materials.

Zidovudine, or Azidothymidine (tradename: Retrovir) is an antiretroviral drug used for the treatment of HIV/AIDS. It acts by stopping HIV from infecting healthy cells, but it can not help cells already infected with the virus [2]. It was the first HIV treatment to be marketed and it is now a widely used generic drug.

Zidovudine is included in the World Health Organisation's "Essential Drugs List" (list of minimum medical needs for a basic healthcare system) [3].

The United States Pharmacopeia (USP) provides worldwide guidance for the chromatographic analysis of Zidovudine [4], which is based on High Performance Liquid Chromatography (HPLC) L1 columns. The implementation of an L1 type column, such as Accucore C18, allowed for the Zidovudine USP analysis time to be reduced from about 30 minutes to less than 2 minutes.



Sample Preparation

A 1000 µg/mL of zidovudine standard solution was prepared in methanol; this solution was then diluted to 100 µg/mL in mobile phase.

Thermo Scientific Column	Part Number
Accucore C18 2.6 µm 100 x 2.1 mm	17126-102130

Thermo Scientific HPLC system

Column temperature	25 °C
Injection volume	1 µL
Flow rate	0.5 mL/min
UV detection	265 nm

Mobile Phase

Mobile phase: water / methanol 4:1 (v/v) .

Consumables	Part Number
Fisher Scientific HPLC grade water	W/0106/17
Fisher Scientific HPLC grade methanol	M/4056/17
NSC Mass Spec Certified 2 mL clear vial with blue bonded PTFE silicone cap MSCERT4000-34W	

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 final run time achieved on the 2.6 µm 100 x 2.1 mm column was 16 times faster than the original run, with zidovudine eluting at 1.6 min (retention time of 26 min on the 250 x 4.0 mm, 5 µm column). Figure 1 shows the chromatogram obtained employing Accucore C18 2.6 µm 100 x 2.1 mm column. The USP acceptance criteria (Tailing factor, $T_f \leq 2.0$, % RSD (for t_r , Efficiency, Peak Area) ≤ 2.0) were met, as demonstrated in Table 1. The statistical assessment is based upon data derived from 6 replicate injections (see Table 1 for mean values).

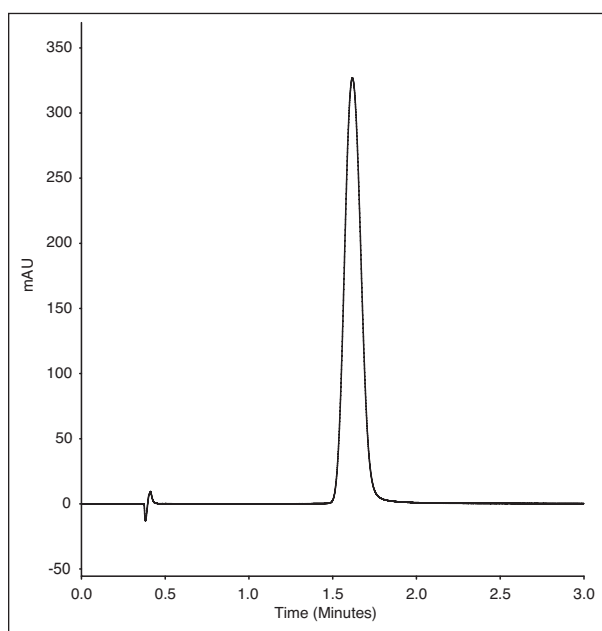


Figure 1: Chromatogram of 100 µg/mL of zidovudine separated on an Accucore C18 2.6 µm 100 x 2.1 mm column

	USP specification	Zidovudine
Asymmetry	< 2.0	1.13
%RSD t_r	< 2.0	0.0
%RSD Peak area	< 2.0	0.34

Table 1: Method precision (%RSD) for zidovudine (data calculated from six replicate injections)

Conclusions

The use of Accucore C18 column, allowed to successfully scale down the USP method for the analysis of zidovudine, in order to increase sample throughput. The analytical results passed the specifications stated in the USP monograph. Accucore C18 columns are therefore an excellent choice for the fast analysis of zidovudine, allowing a high sample throughput.

References

- [1] <http://www.hplctransfer.com/>
- [2] W. V. Caufield; J. T. Stewart; Chromatographia (2001); Vol 54; pp 561-568.
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