Research Article

Development and Validation of New Spectrophotometric Method for the Estimation of Zidovudine in Bulk and Pharmaceutical Dosage Form

Divya K*, Pushpa Latha E and Rekha Rani K

Department of Pharmaceutical Analysis, Creative Educational Society's College of Pharmacy, Chinnatekur, Kurnool – 518218.

ABSTRACT

A simple, accurate, precise, sensitive and a highly selective spectrophotometric method was developed and validated for the estimation of Zidovudine in bulk and pharmaceutical dosage form. The estimation was carried out at maximum wavelength of 281 nm. The linearity was found in the concentration range of 5-50 µg/mL. The correlation coefficient (r^2) was 0.9992. The regression equation was found to be Y = 0.0435C + 0.0205 and % RSD was found to be 2%. The developed method was validated according to ICH guidelines and was found to be accurate and precise. The validation parameters are linearity, accuracy, precision, limit of detection, limit of quantitation, robustness and ruggedness. Thus the proposed method can be successfully applied for the estimation of Zidovudine in bulk and pharmaceutical dosage form.

Keywords: Zidovudine, Validation, ICH guidelines, Spectrophotometric method.

1. INTRODUCTION

Zidovudine (INN) or azidothymidine (AZT) is a nucleoside analog reverse transcriptase inhibitor (NRTI), a type of antiretroviral drug [1-3]. It is a synthetic drug with pyrimidine nucleoside analogue active against HIV-1, AIDS and pre- AIDS. The chemical name of Zidovudine is 1- (3-azide-2, 3-di deoxy-β-D-ribofuranosyl)-5-methyl Pyrimidin-2, 4 (1H, 3H) – dione. Zidovudine also has been referred to as 3′azido-3′-deoxythymidine. It has a molecular formula of $C_{10}H_{13}N_5O_4$ and a molecular weight of 267.24 g/mol.

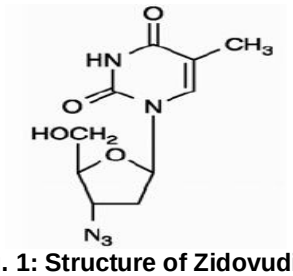

Fig. 1: Structure of Zidovudine

Zidovudine is a white to beige, odorless, crystalline solid and it is soluble in ethanol (95%), sparingly soluble in water. The drug is officially listed in United States of Pharmacopiea [4]. Several analytical methods that have been reported for the estimation of Zidovudine in biological fluids or pharmaceutical formulations include UV-Visible Spectrophotometry [5-6], High Performance Liquid Chromatography [7-12] and HPTLC [13-14]. The objective of the work is to develop a simple, accurate, precise and economic UV spectrophotometric method for the estimation of Zidovudine in bulk and pharmaceutical dosage form. The method is simple, reproducible and statistically valid.

2. MATERIALS AND METHODS

2.1 Materials

Zidovudine was obtained as a gift sample from Hetero drugs Ltd, Hyderabad. Tablets were procured from the local market. Distilled water was used for the preparation of solutions.

2.2 Instrument

Labindia – 3000+ UV / Vis double beam Spectrophotometer (UV Win software) with a fixed slit width (2 nm) and 10 millimeter quartz cell was used to obtain spectrum and absorbance measurement.

2.3 METHOD

2.3.1 Preparation of Stock solutions

Standard Zidovudine 100 mg was weighed and dissolved in 100 ml of water in a 100 ml volumetric flask. The flask was shaken and volume was made up to the mark with water to give a solution containing 1000 µg/ml (stock solution I). From the stock solution I, pipette out 10ml and placed into 100 ml volumetric flask. The volume was made up to mark with distilled water to give a stock solution containing 100 µg/mL (stock solution II).

2.3.2 Selection of analytical concentration ranges

From the standard stock solution II of Zidovudine, appropriate aliquots were pipetted out into 10 ml volumetric flasks and dilutions were made with distilled water to obtain working standard solutions of concentrations from 5 to 50 µg / mL. Absorbance for these solutions were measured at 281nm and the spectra was shown in Figure 2. Values were reported in Table 1.

2.3.3 Calibration curve for the Zidovudine (5 – 50 µg / ml)

Appropriate volume of aliquots from standard Zidovudine stock solution II were transferred to different volumetric flasks of 10 ml capacity. The volume was adjusted to the mark with distilled water to obtain concentrations of 5, 10, 15, 20, 25, 30, 35, 40, 45 and 50 µg / ml. Absorbance spectra of each solution against distilled water as blank were measured at 281 nm and the graphs of absorbance against concentration were plotted and shown in Figure 3. The regression equation and coefficient of determination was determined.

2.3.4 Preparation for Sample Solution

Twenty tablets of Zidovudine were weighed and finely powdered. The powder equivalent to 100 mg of Zidovudine was accurately weighed and transferred to volumetric flask of 100 mL capacity containing 25 mL of the water and sonicated for 5 min. The flask was shaken and volume was made up to the mark with water to give a solution of 1000 µg/ml (stock solution I). The above solution was centrifuged at 2000 rpm for 10 minutes and carefully filtered through Whatmann filter paper (No. 41). From this solution, 10 ml was taken and diluted to 100 ml with distilled water to give a solution of 100 µg/mL (stock solution II) and used for the estimation of Zidovudine. To examine the absence of either positive or negative interference of excipients used in formulation, recovery studies were carried out.

2.3.5 Method validation

Accuracy was determined by recovery studies. The recovery studies were carried out by adding the known amount of standard Zidovudine drug to the sample solution of the tablets. Precision for assay were determined by repeatability, interday, intraday precision for drug (each in three replicate). Ruggedness studies were carried out by changing the analysts. LOD and LOQ were performed and those were values within the limits.

Figure 2 UV Spectra of Zidovudine at 281 nm

Table 1: Results of calibration curve at 281 nm for Zidovudine

S.No	Conc. (mcg/ml)	Absorbance at 281nm
1.	5	0.019
2.	10	0.049
3.	15	0.081
4.	20	0.115
5.	25	0.140
6.	30	0.174
7.	40	0.236
8.	50	0.304

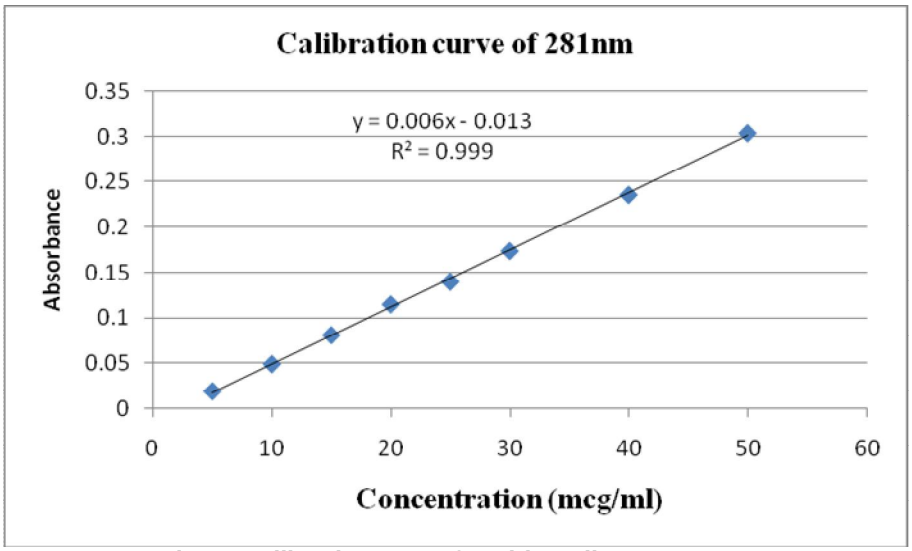

Fig. 3: Calibration curve for Zidovudine at 281 nm

3. RESULTS AND DISCUSSION

Maximum absorption for Zidovudine in UV spectrophotometric method was recorded at 281 nm. The method was validated according to ICH guide lines[15-18]. The optical characteristics such as Beer's law limit, molar absorptivity and other parameters are summarized in Table 2. The results of accuracy, precision and ruggedness studies were shown in Tables 3-6, respectively.

Table 2: Optical Characteristics of Zidovudine

Parameter	UV method
λ_{max} (nm)	281
Beer's law limits (mcg / ml)	5-50
Sandell's sensitivity (mcg / cm^2-0.001 absorbance units)	0.26
Regression equation (Y*)	y = 0.0063x - 0.0137
Slope (b)	0.0063
Intercept (a)	-0.0137
Correlation coefficient(r^2)	0.9994
% RSD**	< 2%
Limit of detection (mcg / ml)	0.127
Limit of quantitation (mcg / ml)	0.386

Table 3: Accuracy results of Zidovudine at 281nm

Brand name	Amount of sample (mcg/ml)	Amount of drug added (mcg/ml)	Amount recovered	%recovery±SD**
Detrol	10	8	17.69	98.27±0.14
Detrol	10	10	19.76	98.83±0.54
Detrol	10	12	21.90	99.54±0.46

**Average of six determinations

Table 4: Precision results of Zidovudine at 281 nm

Conc (mcg/ml)	Interday absorbance Mean±SD	%RSD	Intraday absorbance Mean±SD	%RSD
LQC (10mcg/ml)	0.067±0.0035	1.410	0.0105±0.001446	1.35
MQC (20mcg/ml)	0.106±0.0017	1.189	0.103±0.00238	0.6636
HQC (40mcg/ml)	0.309±0.01616	0.887	0.287±0.01879	1.27

4. CONCLUSION

From the results, it can be concluded that the proposed method for the estimation of Zidovudine is simple, convenient, accurate, sensitive and reproducible. It can be successfully used for routine analysis of the Zidovudine in bulk and pharmaceutical dosage form.

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