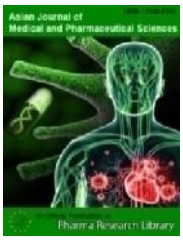
Asian Journal of Medical and Pharmaceutical Sciences

Journal Home Page: www.pharmaresearchlibrary.com/ajmps



Research Article **Open Access**

Simple Validated UV method for Lamivudine in bulk and It's Tablets Dosage form

Asma Khanam S, Ishaq B, Hindustan Abdul Ahad*, Anusha G, Ananda Lakshmi K, Roja G

Balaji College of Pharmacy, Rudrampeta bypass, Anantapur, Andhra Pradesh, India

A B S T R A C T

Lamivudine is prescribed to HIV patients. The present research work is to develop and validate Lamivudine by UV spectrometric method. Simple, accurate, precise and cost efficient spectrophotometric method has been developed for the estimation of Lamivudine in bulk and its tablets dosage form. The optimum condition for the analysis of the drug was established. The maximum wavelength (λmax) was found to be 270nm in water. The mean percentage recovery of Lamivudine was found to be in range of 99.40-101.64%. Beers law was obeyed in the concentration range of 10-200µg/ml. Calibration curve shows a linear relationship between the absorbance and concentration. The line equation y = 0.04x+0.0923 with R^2 = 0.9992 was obtained. Validation was performed as ICH guidelines for linearity, accuracy, precision, LOD & LOQ. The proposed method may be suitable for analysis of Lamivudine in bulk and tablets formulation for routine quality control purposes.

Keywords: Lamivudine tablets, UV spectrophotometric, beers law

A R T I C L E I N F O

CONTENTS

Article History: Received 18 February 2016, **Accepted** 11 March 2016, **Available Online** 19 June 2016

***Corresponding Author**
Hindustan Abdul Ahad
Balaji College of Pharmacy,
Rudrampeta bypass, Anantapur,
Andhra Pradesh, India
Manuscript ID: AJMPS2962

PAPER-QR CODE

Citation: Hindustan Abdul Ahad, *et al.* Simple Validated UV method for Lamivudine in bulk and It's Tablets Dosage form. *A. J. Med. Pharm, Sci.*, 2016, 4(1): 01-05.

1. Introduction

Lamivudine (2′,3′-dideoxy-3′-thiacytidine) (shown in fig.1) commonly called as 3TC is an antiretroviral medication used to prevent and treat HIV/AIDS and used to treat chronic hepatitis B. It is of the nucleoside analog reverse transcriptase inhibitor (NRTI) class. It is on the World Health Organization's List of Essential Medicines, a list of

the most important medication needed in a basic health system [1-5].

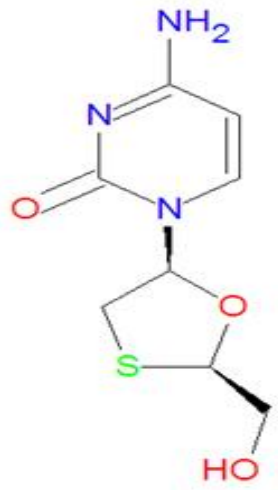


Figure1: Chemical Structure of Lamivudine

2. Experimental

Experimental methods:

An UV–Visible double beam spectrophotometer with 1cm matched quartz cells were used for the spectral and absorbance measurements. All the chemicals and reagents used were of analytical grade and the aqueous solutions were freshly prepared with triple distilled water [6-8].

Preparation of Stock solution

Accurately weighed 10 mg of Lamivudinewas transferred into 100 ml volumetric flask, dissolved in 10ml of water. Take 0.1 ml from above solution and make up to 10ml. Take 0.5 ml from the above solutionand make up to 10 ml to get a concentration of 0.5µg/ml. working standard solution of 1µg/ml for10ml with water. The working standard solutions were daily prepared by diluting stock solution in water [9].

Preparation of test sample

Ten tablets of lamivudine were weighed and powdered. The quantity of the powder equivalent to 10 mg (31.1mg) of lamivudine was transferred in to 10ml of volumetric flask. Water was added up to 10ml and mixed for 5-10 min, filtered the solution and first few ml was discarded. 0.1 ml from the above stock solution was transferred to 10 ml volumetric flask along with water. From the above solution 0.1ml was transferred to 10 ml volumetric flask along with water to get 0.1µg/ml concentration [10].

Solubility Studies

Lamivudine was tested for solubility in various solvents at room temperature [11, 12].

λ_{max} Determination:

The absorbance of the solutions containing Lamivudine at 10µg/ml was determined in the UV range 200-400nm using an appropriate blank [13].

Accuracy

The accuracy of the method was checked by recovery determinations and percentage bias. The determination was done over three concentration levels in triplicate according to the ICH guidelines. The concentration levels selected as QC samples were 5µg/ml, 10µg/ml and 15µg/ml of Lamivudine [14].

Precision

The precision was evaluated on the basis of repeatability and intermediate precision. On at least three occasions, six replicates of each QC sample pool at low, three replicates middle, and six replicates at high concentrations were assayed. Percentage relative standard deviation (%RSD) was calculated [15].

Linearity

The linearity of the described spectrophotometric method was studied in the concentration range 10-100µg/ml for Lamivudine. The calibration curves were constructed by plotting concentration versus intensities. In the overall concentration range examined, the linearity was evaluated by linear regression analysis that was calculated by the least square regression method [16].

Limit of Detection

The detection limit of an individual analytical procedure is the lowest amount of analyte in a sample which can be detected but not necessarily quantitated as an exact value [17].

Limit of Quantification

The Quantitation limit of an individual analytical procedure is the lowest amount of analyte in a sample which can be quantitatively determined with suitable precision and accuracy [18].

Degradation studies

The degradation studies were performed for the Lamivudine in water [19].

3. Results and Discussion

Lamivudine was found to be soluble in Water, Benzene, Glacial acetic acid, Sodium hydroxide, hydrochloric acid and acetone at room temperature. The λ max was found to be 270nm. The λ_{max} of Lamivudine pure drug and in tablets were shown in fig 2 and 3 respectively. The Molar Absorptivity was found to be 7.5 x 10^{-6} by using the formula (e= A / c l), (A= absorbance, c = sample concentration in moles/liter &l = length of light path through the sample in cm). The Sandell's sensitivity (Sandell's sensitivity = mol. wt / molar absorvitity) was found to be 2.96 X 10^{7}µg cm^{-2}.The Accuracy results were shown in table 1.

The precision was carried out for 10µg/ml. The intraday precision for 10µg/ml was found to be 1.493% RSD, while it gave a value of 1.49366% RSD at interday (intermediate precision). The accuracy of the method was calculated as percent bias. The Intraday Precision and Inter-day precision of Lamivudine tablets were shown in table 2 and 3 respectively.

The spectrum of standard solution of Lamivudine in water was given in Fig. 2 and 3.The λ_{max} was found to be 270 nm. The calibration curve was prepared by plotting concentration (in µg/ml) on the abscissa and absorbance in ordinate axis in the range of 10-100µg/ml. Good linearity was obtained in the concentration range considered. The linearity equation was developed by using least square

regression analysis. The Limit of detection and Limit of quantitation was calculated according to the ICH guidelines. The Beer-Lamberts limit was found to be10-100µg/ml (table 4). Calibration curve of Lamivudine was plotted (shown in fig. 4) and the linearity, regression, LOD and LOQ were shown in table 5. The LOD was found to be0.675µg/ml and the LOQ was found to be 2.25µg/ml. The acid, base, heat and peroxide degradation spectrum of Lamivudine tablets were summarized in table 6.

Table 2: Intraday Precision of Lamivudine tablets

S. No	Conc. (µg/ml)	Absorbance		
		Day-I	Day-II	Day-III
1	10	0.548	0.523	0.509
2		0.544	0.528	0.505
3		0.547	0.521	0.513
4		0.566	0.536	0.517
5		0.545	0.539	0.514
6		0.543	0.534	0.528
Average		0.549	0.530	0.514
SD		0.009	0.007	0.008
% RSD		1.569	1.378	1.534

Table 3: Inter-day precision of Lamivudine tablets

S. No	Conc. (µg/ml)	Absorbance		
		Day-I	Day-II	Day-III
1	10	0.548	0.523	0.509
2		0.544	0.528	0.505
3		0.547	0.521	0.513
4		0.566	0.536	0.517
5		0.545	0.539	0.514
6		0.543	0.534	0.528
Average		0.549	0.530	0.514
SD		0.009	0.007	0.008
% RSD		1.569	1.378	1.534
Limit% RSD must be less than 2%.				

Table 4: Calibration (Linearity) of Lamivudine tablets

Concentration (µg/ml)	Absorbance
1	0.129±0.001
5	0.287±0.001
10	0.507±0.001
15	0.691±0.002
20	0.887±0.002
Values are in mean ±S.D: number of trials (n)=3	

Table 5: Slope, Linearity, LOD and LOQ of Lamivudine tablets

Slope	Linearity (R^2)	LOD (µg/ml)	LOQ (µg/ml)
0.04x+0.0923	0.9992	0.675	2.25

Table 6: Degradation studies of Lamivudine tablets

Parameter	Result
Acid degradation	0.545±0.001
Base degradation	0.464±0.001
Peroxide degradation	0.543±0.001
Heat degradation	0.523±0.001
Values in mean±S.D; N=3	

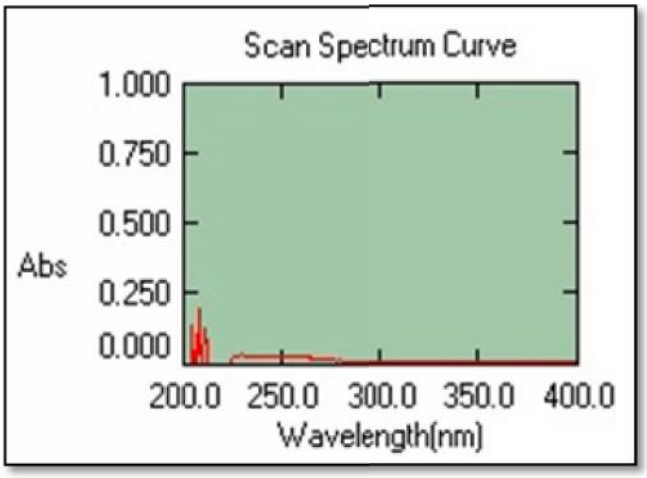


Figure 2: λ_{max} of Lamivudine pure drug

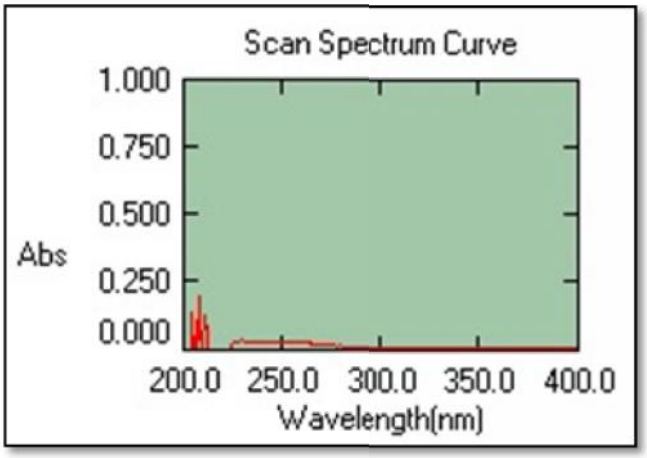


Figure 3: λ_{max} of Lamivudine tablet powder

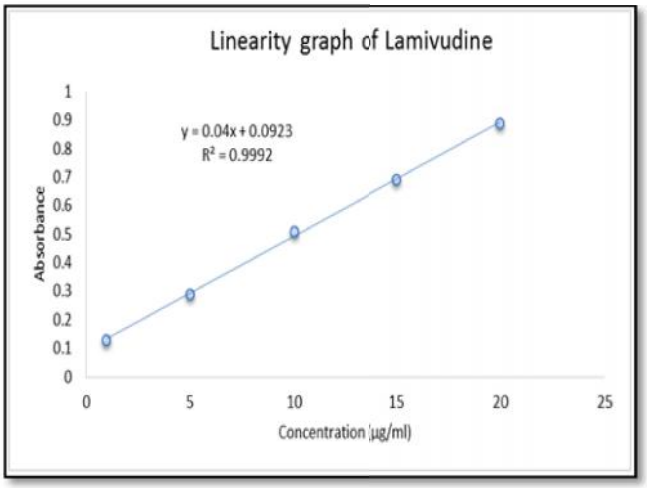


Figure 4: Calibration curve of Lamivudine

Table 1: Accuracy results of Lamivudine tablets

Conc. Level	Sample Weight (mg)	Absorbance	Amount added	Amount found	% Recovery	Mean % Recovery
50%	15.55	0.275	4.969	4.964	99.888	99.403
		0.276	4.969	4.982	100.251	
		0.274	4.969	4.946	99.524	
		0.272	4.969	4.910	98.798	
		0.271	4.969	4.892	98.435	
		0.274	4.969	4.946	99.524	
100%	31.1	0.537	9.939	9.693	97.527	98.616
		0.526	9.939	9.495	95.529	
		0.566	9.939	10.217	102.794	
150%	46.65	0.835	14.908	15.072	101.098	101.643
		0.836	14.908	15.090	101.220	
		0.866	14.908	15.632	104.852	
		0.836	14.908	15.090	101.220	
		0.826	14.908	14.910	100.009	
		0.838	14.908	15.126	101.462	

4. Conclusion

In this study simple, fast and reliable UV spectrophotometric method was developed and validated for the determination of Lamivudine in tablet formulation. The method was applied directly to the analysis of pharmaceutical dosage forms without the need for separation such as extraction steps prior to the drug analysis. As these proposed methods have the lowest LOD value and wider linear range so these methods were more sensitive methods. From the results obtained, we concluded that the suggested methods showed high sensitivity, accuracy, reproducibility and specificity. Moreover, these methods were simple and inexpensive and they can be employed for the routine quality control analysis of Lamivudine in pharmaceutical formulations.

5. References

[1] Goodman, Gileman. The Pharmacological Basis of Therapeutics. In: Joel GH, editor. 11th ed. Mc-Graw Hill: Medical publishers Div; **2006**. p. 1288.

[2] US Pharmacopoeia. Asian Edition 30. United States Pharmacopoeial Convention, Inc: **2007**. p. 2447.

[3] Indian Pharmacopoeia. Vol. 2. Government of India: Controller of Publications; 2. Government of India Ministry of Health & Family Welfare; p. 1276.

[4] HMSO. Cambridge: International edition; Stationery Office (Great Britain). British Pharmacopoeia; Vol. 2, 2007, p. 1216.

[5] Maryadel JO. The Merck Index. In: Maryadel JO, editor. 14th ed. New Jersey: Merck Research Laboratories; **2006**.

[6] Shalini S, Shanooja VP, AbdulJameel S, Basima, Harilal KK, Harish R, et al. Application of UV-spectrophotometric methods for estimation of lamivudine in tablets. Dig J Nanomater Biol Struct. **2009**; 4: p. 357–60.

[7] Basavaiah K, Somashekar BC. Titrimetric and spectrophotometric determination of lamivudine in pharmaceuticals. Indian J Chem Technol. **2009**, 13: p.7–12.

[8] Babu CJ, Kumar GV. Validated RP- HPLC method for the quantification of Lamivudine in bulk and tablet dosage form. Int J Pharm Tech Res. **2009**;1: p.1721–4.

[9] Baig MV, Kapse GS, Raju SA. Spectrophotometric Determination of Lamivudine. Asian J Chem. **2001**; 13: p.185–9.

[10] Mohammed Ishaq B, Muneer S, Hindustan Abdul Ahad,A Simple UV Spectroscopic Method for the Determination of Ritonavir inBulk and Tablets, JPBMAL, 2 (2), **2014**, p.118-122.

[11] U.S. Food and Drug Administration Guidance for Industry, ICH Q3B, Impurities in New Drug Products, **2006**.

[12] U.S. Food and Drug Administration Guidance for Industry, ICH Q3C, Impurities.

[13] Residual Solvents, 1997.Solli, Chou, Jalali B et al. "Amplified wavelength–time transformation for real-time spectroscopy," Nature Photonics, **2008**, 2, p.48-51.

[14] Ishaq BM, Hindustan Abdul Ahad, UV Spectroscopic Method for Estimation of Enalapril Maleate in Bulk andin its Tablets, AMCL, 1(1):**2014**, p.31–35

[15] Hoetelmans RM, Profijit M, Mennhorst PL, Mulder JW, Beijnen JH. Quantitative determination of (−)-2′-deoxy-3′-thiacytidine (lamivudine) in human plasma, saliva and cerebrospinal fluid by high-performance liquid chromatography with ultraviolet detection. J. Chromatogr. B Biomed. Sci. Appl., **1998**, 713: p.387–394.

[16] Moore JD, Valette G, Darque A, Zhou XJ, Sommadossi JP. Simultaneous quantitation of the 5'-triphosphate metabolites of zidovudine, lamivudine, and stavudine in peripheral mononuclear blood cells of HIV infected patients

by high-performance liquid chromatography tandem mass spectrometry. J. Am. Soc. Mass Spectrom., **2000**, 11: p.1134–1143.

[17] Kirkbright GF. Development and publication of new spectrophotometric methods of analysis. Talanta, **1966**, p.1-14.

[18] Willard HH, Merrit LL (Jr), Dean JA, Settle FA (Jr). Instrumental methods of analysis. 6th ed. New Delhi: CBS Publishers and Distributors; **1986**, p.1-15.

[19] Sarma GN, Sastry CS, Sastri CK. Simple Oxidimetric methods for determination of Stavudine or lamivudine. Asian J Chem. **2002**; 14: p.683–90.