International Journal of
Medicinal Chemistry & Analysis

www.ijmca.com

e ISSN 2249 - 7587
Print ISSN 2249 - 7595

SPECTROPHOTOMETRIC METHOD FOR DEGRADATION STUDY OF LAMIVUDINE

Gouthami K*, Anusha Priyadarshini K, Nikhila Soundarya A, Pushpa Latha E

Department of Pharmaceutical Analysis and Quality Assurance, Creative Educational Society's College of Pharmacy, Chinnatekur, Kurnool – 518218, Andhra Pradesh, India.

ABSTRACT

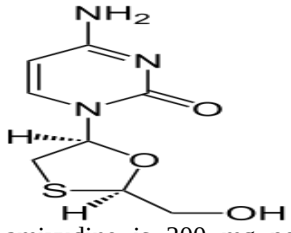
Spectrophotometric method for degradation study of Lamivudine was described. The ultra violet spectrum of acidic, basic and oxidative degraded product was found to substantial difference from pure drug. The extent of degradation can be calculated by comparing the decrease in absorbance at selective wavelength. The UV spectrum of Lamivudine showed maximum absorbance at 270nm. The solution in NaOH showed a decrease in absorbance at 270 nm. So the decrease in absorbance at 270nm was used as measure of extent of degradation in NaOH. In case of NaOH degradation also decrease of absorbance is seen due to degradation but in comparison to NaOH degradation is lesser. In the basic degradation study it was found that Lamivudine is more sensitive to alkaline hydrolysis.

Keywords: Lamivudine, UV spectrophotometry, Degradation.

INTRODUCTION

Lamivudine is antiretroviral drug and acts by blocking reverse transcriptase [1]. Chemically Lamivudine is 4-amino-1-[(2R,5S)-2-(hydroxymethyl)-1,3-oxathiolan-5-yl]-1,2 dihydropyrimidin-2-one [2]. It has a molecular formula of $C_8H_{11}N_3O_3S$ and a molecular weight of 229.26 g / mol and its structure was given in Fig 1.

Fig 1. Chemical Structure of Lamivudine



The dose of Lamivudine is 300 mg per day. Several combinations of Lamivudine with other antiretroviral drugs are available in the market for treatment of HIV infected patients [3]. Literature survey revealed, few analytical methods which include simultaneous determination of Lamivudine and Stavudine in human serum using HPLC with tandem mass spectrometry [4], development and validation of normal phase HPTLC method for analysis of Lamivudine, Stavudine and Nevirapine in fixed dosed combination tablet [5], simultaneous determination of HIV nucleoside analogues of reverse transcriptase inhibitors Lamivudine, Didanosine, Stavudine, Zidovudine and Abacavir in human plasma using reverse phase high performance liquid chromatography [6]. Simultaneous determination of Lamivudine, Stavudine and Nevirapine in antiretroviral fixed dose combination by high performance liquid chromatography [7], Validation of high-performance liquid chromatography methods for determination of Zidovudine, Stavudine, Lamivudine and Indinavir in human plasma [8]. Lamivudine is not official in IP, BP and USP. The present work deals with estimation of Lamivudine in tablets by UV-Spectrophotometry and first order derivative [9].

MATERIALS AND METHODS

Drug and Chemicals

All chemicals used were of A.R Grade from S.D.Fine – Chem, Merck, Fischer scientific, and spectrochem, Mumbai. Authentic drug sample of

Corresponding Author: - **Gouthami K** Email:gouthami.mpharm@gmail.com

Lamivudine was obtained as a gift sample by Hetero drugs Ltd., Hyderabad.

Instrument

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

Preparation of stock solution

Standard stock solution was prepared by dissolving accurately weighed 100mg of Lamivudine in water and the volume was made up to 100ml with water in a 100ml volumetric flask (1000 mcg/ml). 10ml of above stock solution-1 was diluted to 100ml with water (stock solution-2,100mcg/ml).1ml of stock solution-2 was taken in 10ml standard flask diluted to 10ml with water to get the concentration 10mcg/ml. The absorbance of resulting solution was measured against respective blank solution in the UV region of 200-400nm, which shows maximum absorbance at 270nm.

Forced degradation Study

Alkaline Degradation Study: Alkaline degradation was done against 0.1N NaOH.

Accurately weighed 100mg of Lamivudine was dissolved in 100ml volumetric flask with 100ml water. From the above stock solution, 1ml is taken and is made up to 10ml with 0.1N NaOH in 10ml volumetric flask. From that solution, 1ml is taken and made up to 10ml in 10ml volumetric flask at time intervals of 1hr, 2hr, 24hr and 48hrs.

Acidic Degradation study: Acidic degradation was done against 0.1N HCl.

Accurately weighed 100mg of Lamivudine was dissolved in 100ml of water in 100ml of volumetric flask. From the above solution 1ml is taken out and it is made up with 0.1N HCl in 10ml volumetric flask. From the solution 1ml is taken and made up to 10ml volumetric flask at time intervals of 1hr, 2hr, 24hr and 48hrs.

Oxidative Degradation: Oxidative degradation was done against 0.3% H_2O_2

Accurately weighed 100mg of Lamivudine was dissolved in 100ml of water in 100ml of volumetric flask. From that solution 1ml is taken out and it is taken out and it is making up to 10ml with 0.3%H_2O_2 in 10ml volumetric flask. From that solution 1ml is taken and made up to 10ml in 10ml volumetric flask at time intervals of 1hr, 2hr, 24hr and 48hr. The absorbance of above solutions was noted.

RESULTS

Lamivudine showed maximum absorbance at 270nm.The sample in NaOH showed decrease in the absorbance at 270nm and was used as the measure of extent of degradation in NaOH.In case of HCl degradation also, decrease of absorbance is seen due to degradation but in comparison to NaOH degradation is lesser. Absorbance is seen at 270nm and there is no peak observed in NaOH .The overlain zero order spectrum of Lamivudine, NaOH degraded and HCl degraded respectively showed in figure.

Table 1. Degradation of Lamivudine in acidic condition [0.1N HCL]

S .No	Time	Absorbance
1	0hr	0.620
2	1hr	0.553
3	2hr	0.503
4	48hr	0.496

Table 2. Degradation of Lamivudine in alkali condition [0.1N NAOH]

S .No	Time	Absorbance
1	0 hr	0.620
2	1hr	0.388
3	2hr	0.359
4	48hr	0.329

Table 3. Degradation of Lamivudine in oxidative stress condition (0.3%H_2O_2)

S .No	Time	Absorbance
1	0hr	0.620
2	1hr	0. 648
3	2hr	0.629
4	48hr	0.663

Fig 2. Spectra of Lamivudine at 270 nm

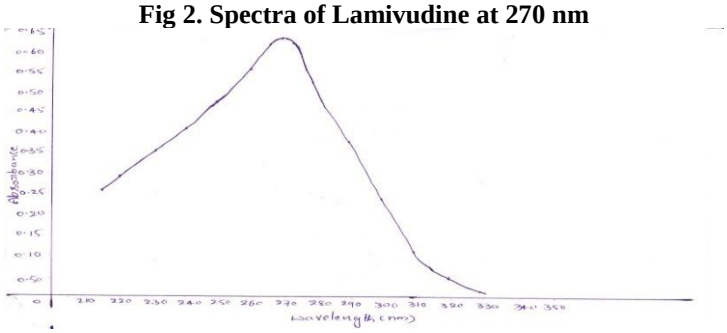
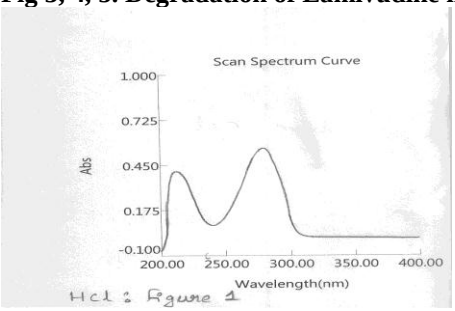
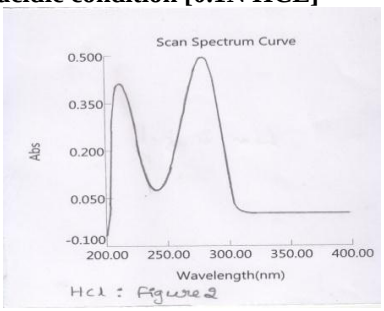


Fig 3, 4, 5. Degradation of Lamivudine in acidic condition [0.1N HCL]





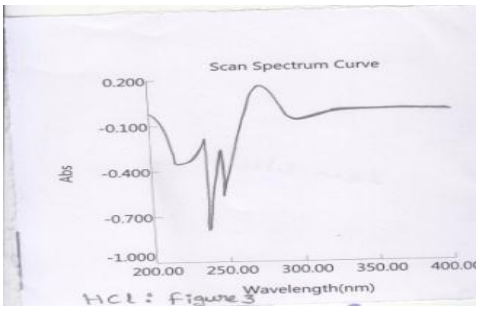
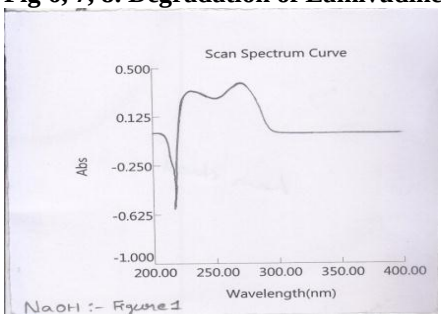
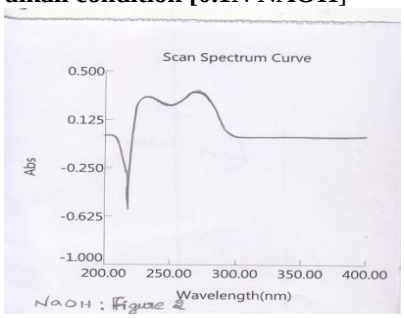


Fig 6, 7, 8. Degradation of Lamivudine in alkali condition [0.1N NAOH]





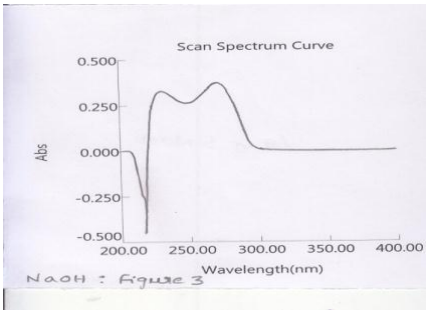
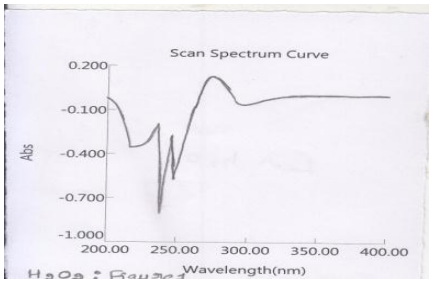
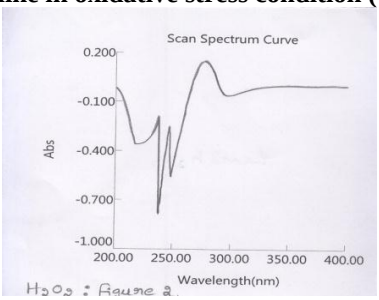
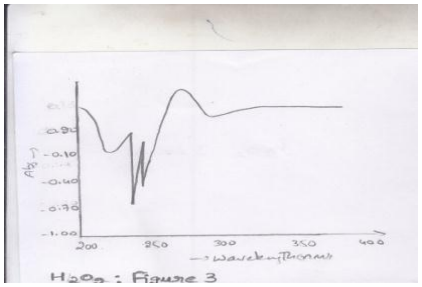


Fig 9, 10, and 11. Degradation of Lamivudine in oxidative stress condition (0.3%H_2O_2)







DISCUSSION

The ultra violet spectrum of both acidic and basic degraded products was found to be substantially different from pure drug. The extent of degradation can be calculated by comparing the decrease in absorbance at selective wavelength. The solution which was degraded in NaOH and HCl showed a decrease in the absorbance at 270nm and increase in absorbance at 231nm was measure of extent of degradation in NaOH and NaOH and no degradation has been observed in oxidative stress condition. We can observe the degradation of Lamivudine with its respective solution in figures. The degradation for each concentration was calculated by comparing the decrease in absorbance with untreated drug solution. In the basic degradation study, it is found that Lamivudine is more sensitive to alkaline condition.

CONCLUSION

In the basic degradation study, it is found that Lamivudine is more sensitive to alkaline hydrolysis. The degradation is somewhat slower in acidic conditions of drug is degraded if treated with 0.1N hydrochloric acid. Lamivudine didn't show any degradation in H_2O_2. The proposed method could be applied for routine analysis in quality control laboratories.

REFERENCES

1. Sean CS. The complete drug reference. In: Sean CS, editor. 35th edn. Pharmaceutical Press, London. 2009.
2. Maryadele J, Neil O. The Merck Index,14th edn, Published by Merck and Co, White House Station, NJ, USA, 2006, 5146.
3. Gallant JE, Dejesus E, Arribas JR *et al.*, Tenofovir DF, Emtricitabine, and Efavirenz vs. Zidovudine, Lamivudine, and Efavirenz for HIV. *N.Engl.J.Med,* 354(3), 2006, 251-260.

4. Kenney KB, Wring SA, Carr RM, Wells GB. Simultaneous determination of Lamivudine and Stavudine in human serum using HPLC with tandem mass spectrometry. *Journal of Pharmaceutical and Biomedical Analysis*, 22, 2000, 867-838
5. Shewiyo DH, Kaale E, Ugullum C, Sigonda MN, Smeyers-Verbeke J, Vander HY. Development and validation of normal phase HPTLC method for analysis of Lamivudine, Stavudine and Nevirapine in fixed dosed combination tablet. *Journal of Pharmaceutical and biomedical analysis*, 31, 2011, 445-450.
6. Verweij VW, Aarnoutse RE, Burger DM. Simultaneous determination of HIV nucleoside analogues of reverse transcriptase inhibitors Lamivudine, Didanosine, Stavudine, Zidovudine and Abacavir in human plasma using reverse phase high performance liquid chromatography. *Journal of Chromatography B*, 816, 2005, 121-129.
7. Kapoor N, Khandavilli S, Panchaghula R. Simultaneous determination of Lamivudine, Stavudine and Nevirapine in antiretroviral fixed dose combination by high performance liquid chromatography. *Analytica Chimica Acta*, 570, 2006, 41-45.
8. Tarinas A, Tápanes RD, Ferrer G, Pérez J. Validation of high-performance liquid chromatography methods for determination of Zidovudine, Stavudine, Lamivudine and Indinavir in human plasma. *Farmacia Hospitalaria*, 31(4), 2007, 243-247.
9. Beckett AH, Stenlake JB. Practical Pharmaceutical chemistry, 4th edn., part II, S.K. Jain for CBS publisher and Distributors. New Delhi, 1998, 275-300.