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Spectrophotometric Estimation of Abacavir Sulphate in Pharmaceutical Formulations

N. APPALA RAJU, J. VENKATESWARA RAO*,
K. VANITHA PRAKASH and K. MUKKANTI[#]

Department of Pharmaceutical Chemistry, Sultan-UI-Uloom College of Pharmacy,
Mount Pleasant, Road No. 3, Banjara Hills, Hyderabad-500 034.

[#]Institute of Science and Technology, Jawaharlal Nehru Technological University,
Kukatpally, Hyderabad -500072, India.

jvrao1963@yahoo.co.in

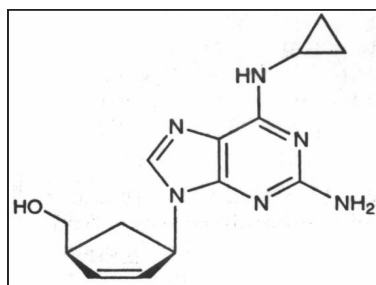
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Abstract: Two simple, accurate, rapid and sensitive methods (A and B) have been developed for the estimation of abacavir sulphate in its pharmaceutical dosage form. The method A and B are based on the formation of chloroform extractable complex of abacavir sulphate with bromophenol blue (method A) and bromocresol green (method B), which shows absorbance maxima at 460 nm and 469 nm respectively. The absorbance-concentration plot is linear over the range of 1-10 mcg/mL for method A and B respectively. Results of analysis for all the methods were validated statistically and by recovery studies. The proposed methods are economical and sensitive for the estimation of abacavir sulphate in bulk drug and in its tablet dosage form.

Keywords: Ultraviolet-Visible Spectrophotometry, Abacavir sulphate, Bromophenol Blue (BPB), Bromocresol Green (BCG).

Introduction

Abacavir sulphate¹ is chemically {(1S, 4R)-4-[2-Amino-6-(cyclopropylamino)-9H-purin-9-yl]-2-cyclopentene-1-methanol}. It is a nucleoside reverse transcriptase inhibitor with antiretroviral activity against HIV. It is administered alone or in combination therapy with other antiretrovirals. Survey of literature reveals that the drug is determined by using High Performance Liquid Chromatography²⁻⁴ only. No spectrophotometric methods are reported. The present study describes simple, sensitive, accurate, rapid and economical spectrophotometric methods for the estimation of abacavir sulphate in bulk & its tablet dosage forms.



Structure of Abacavir

Experimental

Instrument

Elico Ultraviolet-Visible double beam spectrophotometer SL-164 with 1 cm matched quartz cells was used for all spectral measurements.

Reagents

All the chemicals used were of analytical reagent grade. All the solutions were freshly prepared.

1. Acid phthalate buffer pH 3
2. Bromophenol Blue (0.1%)
3. Acid phthalate buffer pH 2.4
4. Bromocresol Green (0.1%).
5. Chloroform AR grade.

Procedure

A standard stock solution containing 1 mg/mL was prepared by dissolving 100 mg of abacavir sulphate in 100 mL of distilled water for method A and B. From this, a working standard solution containing 20 µg/mL was prepared for method A and B.

Assay procedure

Method A

Aliquots of standard drug solution of abacavir sulphate 0.5 – 5.0 mL (20 mcg/mL) were taken and transferred into a series of 125 mL of separating funnels. To each funnel 2 mL of 0.1% BPB was added. Reaction mixture was shaken gently for 5 min. Then 10 mL of chloroform was added to each of them. The contents were shaken thoroughly for 5 min and allowed to stand, so as to separate the aqueous and chloroform layer. Colored chloroform layer was separated out and absorbance was measured at 460 nm against reagent blank. Calibration curve was plotted from absorbance values so obtained

Method B

Aliquots of standard drug solution of abacavir sulphate 0.5 – 5.0 mL (20 mcg/mL) were taken and transferred into a series of 125 mL of separating funnels. To each funnel 2 mL of 0.1% BCG was added. Reaction mixture was shaken gently for 5 min. Then 10 mL of chloroform was added to each of them. The contents are shaken thoroughly for 5 min and allowed to stand, so as to separate the aqueous and chloroform layer. Colored chloroform layer was separated out and absorbance was measured at 469 nm against reagent blank. Calibration curve was prepared from absorbance values so obtained

Preparation of sample solution

Twenty tablets of abacavir sulphate (Abamune, 300 mg, Cipla) were accurately weighed and powdered. Tablet powder equivalent to 100 mg of abacavir sulphate was dissolved in 50 mL of distilled water, sonicated for 15 mins, filtered and washed with distilled water. The filtrate and washings were combined and the final volume was made to 100 mL with distilled water. The solution was suitably diluted and analyzed as given under the assay procedure for bulk samples. The results are represented in Table 1. None of the excipients usually employed in the formulation of tablets interfered in the analysis of abacavir, by the proposed methods.

Recovery Studies

To ensure the accuracy and reproducibility of the results obtained, known amounts of pure drug was added to the previously analysed formulated samples and these samples were reanalyzed by the proposed method and also performed recovery experiments. The percentage recoveries thus obtained were given in Table 1.

Table 1. Assay and recovery of abacavir sulphate in tablet dosage form.

Tablet formulation	Labeled Amount, mg	Amount Obtained by proposed method, mg*		** % Recovery by the Proposed method	
		Method A.	Method B	Method A	Method B
1	300	297.9	301.5	99.3	100.5
2	300	298.5	302.3	99.4	101.2
3	300	301.3	297.3	100.4	99.1

Average of three determinations **After spiking the sample.

Results and Discussion

The optimum conditions were established by varying one parameter at a time and keeping the others fixed and observing the effect on absorbance of chromogen. In the present work method A and B have been developed for the estimation of abacavir sulphate from tablet formulation. The developed methods A and B are based on formation of chloroform extractable colored complexes with bromophenol blue and bromocresol green respectively. The conditions required for the formation of colored complexes were optimized. Statistical analysis was carried out and the results were found to be satisfactory. Relative standard deviation values were low that indicates the reproducibility of the proposed methods. Recovery studies were close to 100 % that indicates the accuracy and precision of the proposed methods.

The optical characteristics such as absorption maxima, Beer's law limits, molar absorptivity and Sand ell's sensitivity are presented in Table 2.

Table 2. Optical characteristics and precision data

Parameters	Method A	Method B
λ_{max} , nm	460	469
Beer's law limits, mcg/mL	1-10	1-10
Molar absorptivity, L/mol.cm	1.174×10^3	8.877×10^3
Sand ell's sensitivity, micrograms/cm ² /0.001 absorbance unit	0.243	0.0322
Regression Equation* (Y) Slope (m) Intercept (c)	0.005-0.0046	0.006-0.419
Correlation Coefficient(r)	0.9999	0.9998
Precision (%Relative Standard Deviation)	0.264	0.502
Standard error of mean	0.019	0.0037

* $Y=mx+c$, where X is the concentration in micrograms/ml and Y is absorbance unit.

The regression analysis using the method of least squares was made for slope (m), intercept (b) and correlation obtained from different concentrations and the results are summarized in Table 2.

Conclusion

The proposed methods are simple, sensitive, accurate and economical for the routine estimation of abacavir sulphate in bulk and in its tablet dosage form.

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