Determination of Acebrophylline in bulk and pharmaceutical formulation by UV spectrophotometer.

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Abstract

A simple, sensitive and accurate spectrophotometric method has been developed for the determination of Acebrophylline in bulk drug and capsule. The λ max of the Acebrophyllline was found to be 274 nm. The method showed high sensitivity with linearity in the range of 2 to 20µg/ml, coefficient regression was found to be 0.9994. The regression of the curve was found to be Y = 0.0187X - 0.0062. Percent relative standard deviation value is below 2.0 for intraday and interday precision indicated that method is highly precised. The percentage recovery value (average 100.1458 percentages), indicated the accuracy and specificity of the method. The proposed method will be suitable for the analysis of Acebrophylline in bulk and pharmaceutical formulation.

Key Words

Acebrophylline, UV visible spectrophotometer.

Introduction

Acebrophylline,1,2,3,6-tetrahydro-1,3-dimethyl-2,6-dioxo-7H-Purine-7-aceticacid compound with trans-4-[[(2-amino-3,5 dibromophenyl)methyl] amino] cyclohexanol is used in bronchial asthma and pulmonary diseases[1]. Literature survey reveals that very few methods are available for estimation of Acebrophylline in bulk drug and its pharmaceutical formulation by UV spectrophotometry[2]. A new UV spectrophotometric method based on single wavelength analysis[3] of Acebrophylline at max 274 nm was done.
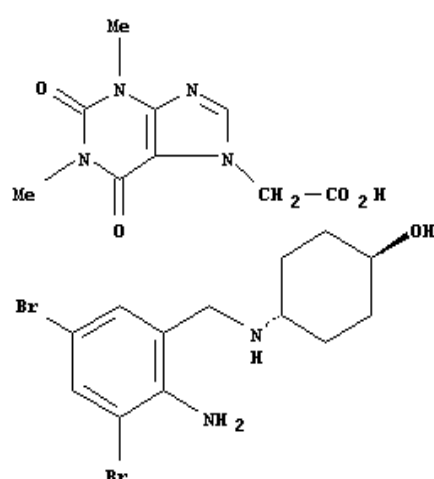

Figure 1: Structure of Acebrophylline.

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Chemicals & reagents

Acebrophylline reference standard was kindly provided by AMI LIFESCIENCE PVT. LTD.82/B, ECP, Canal Rd.Karkhadi, Padra, Baroda. Analytical grade ethanol was purchased from Merck Chemicals, India.

Instrument

All the measurements were made using JASCO UV-V-550 UV Visible Spectrophotometer with 1 cm matched quartz cells.

Methods

Selection of wavelength

In order to ascertain the wavelength of maximum absorption (λmax) of the drug, different solutions of the drugs (10µg/ml) in ethanol were scanned using spectrophotometer within the wavelength range of 200 – 380 nm against ethanol as blank. The resulting spectrum is shown in figure 2 and the absorption curve showed characteristic absorption maxima at 274 nm for Acebrophylline.

Preparation of standard solution for calibration curve

10 mg Acebrophylline was accurately weighed and transferred to 100 ml volumetric flask and was dissolved properly and diluted up to the mark with ethanol to produce a stock solution of 100µg/ml. Then 2ml of this solution was diluted to 10ml with ethanol to give 2µg/ml, similarly concentrations of 4µg/ml, 6µg/ml 8µg/mg,10µg/ml,12µg/mg 14µg/mg

16μg/mg,18μg/mg and 20μg/ml where prepared which were used for the construction of calibration curve (figure 3).

Validation of method[4]

The proposed method was validated according to ICH guidelines for linearity, accuracy, precision, LOD and LOQ.

Linearity

The linearity of the method was evaluated by triplicate analysis of standard solutions (n=3). The plot of absorbance Vs respective concentration (Fig2) of Acebrophylline was found to be linear in the range of 2mg/ml- 20mg/ml. Beer's law was obeyed over this concentration range. The regression equation was found to be y= 0.0187x - 0.0062and the correlation coefficient (r) of the standard curve was found to be 0.9994 (Table no 1).

Precision

The amounts of Acebrophylline was found by the number of replicates of both pharmaceutical preparations (n=3) performed by repeatability (intraday) and intermediate precision (inter-day) and reported as RSD percent relative standard deviation. For this, 10 μg/ml concentration solutions were measured three times in day and same was measured in next three days. The percent relative standard deviation was calculated.

Preparation of sample solution

Twenty tablets were weighed accurately and powdered. Tablet powder equivalent to 10mg of Acebrophylline was weighed and transferred to a 100ml volumetric flask. About 40ml of ethanol was added and sonicated for 5 min for complete dissolution of drugs, the volume was made up to the mark with the same solvent and then the above solution was filtered through Whatmann filter paper. Now After suitable dilution, the absorbance of final sample was recorded against the blank at 274 nm. All determinations were conducted in triplicate.

Recovery study

Recovery study was performed to judge the accuracy of the method. Each level was repeated three times (n = 3). Recovery study was carried out by adding a known quantity of pure drug to the preanalyzed formulation and the proposed method was followed. From the amount of drug found, percentage recovery was calculated (table no 2). Recovery study was carried out at three levels 80%, 100% and 120% for the formulation concentration of 10μg/ml.

LOD & LOQ

LOD (k = 3.3) and LOQ (k = 10) of the method were established according to ICH definitions. LOD and LOQ of method are reported in Table. In this study, LOD and LOQ were based on the standard deviation of the response and the

Slope of the corresponding curve using the following equations-

LOD = 3.3 SD/S;

LOQ = 10 SD/S

Where SD is the standard deviation of the absorbance of the sample and S is the slope of the calibrations curve.

Result and Discussion

The λ max of the Acebrophylline was found to be 274 nm. From the optical characteristics (Table) of the proposed method, it was found that Acebrophylline obeys linearity within the concentration range of 2 to 20 μg/ml and coefficient correlation was found to be 0.9994. The regression of the curve was at Y = 0.0187X + 0.0062 .The detection and quantization limits as LOD (k=3.3) and LOQ (k=10) were calculated and these were found to be 0.14117μg/ml and 0.42780 μg/ml respectively. The percentage recovery value (table) indicated the accuracy and specificity of the method. The proposed method was also applied for the assay of Acebrophylline in tablet formulation (in triplicate) and the results as tabulated in Table. The results obtained were in good agreement with the label claims.

Conclusion

The proposed method is simple, sensitive and reliable with good precision and accuracy. The proposed method is specific while estimating the commercial formulations without interference of excipients and other additives. Hence, this method can be used for the routine determination of Acebrophylline in pure samples and pharmaceutical formulations.

References

1. Beulcke G. Acebrophylline and theophylline in the treatment of bronchial asthma. Anopencomparative, randomizedstudy. Acta Therapeutica 1995;21(2):1011.
2. D. Saraswathi, J. Priyadharisini, V. Niraimathi And A. Jerad Suresh Spectrophotometric Estimation Of Acebrophylline In Bulk And

Capsule Formulation, Int. J. Chem. Sci.: 8(2), 2010, 973-976.

3. A.H. Beckett and J. B. Stenlake, Practical Pharmaceutical Chemistry, 4th Edition, Part Two, CBS Publishers and Distributors, New Delhi, (2007).

4. Validation of Analytical Procedures: Text and Methodology, Proceedings of International Conference on Harmonization (ICH). Geneva, 2005.
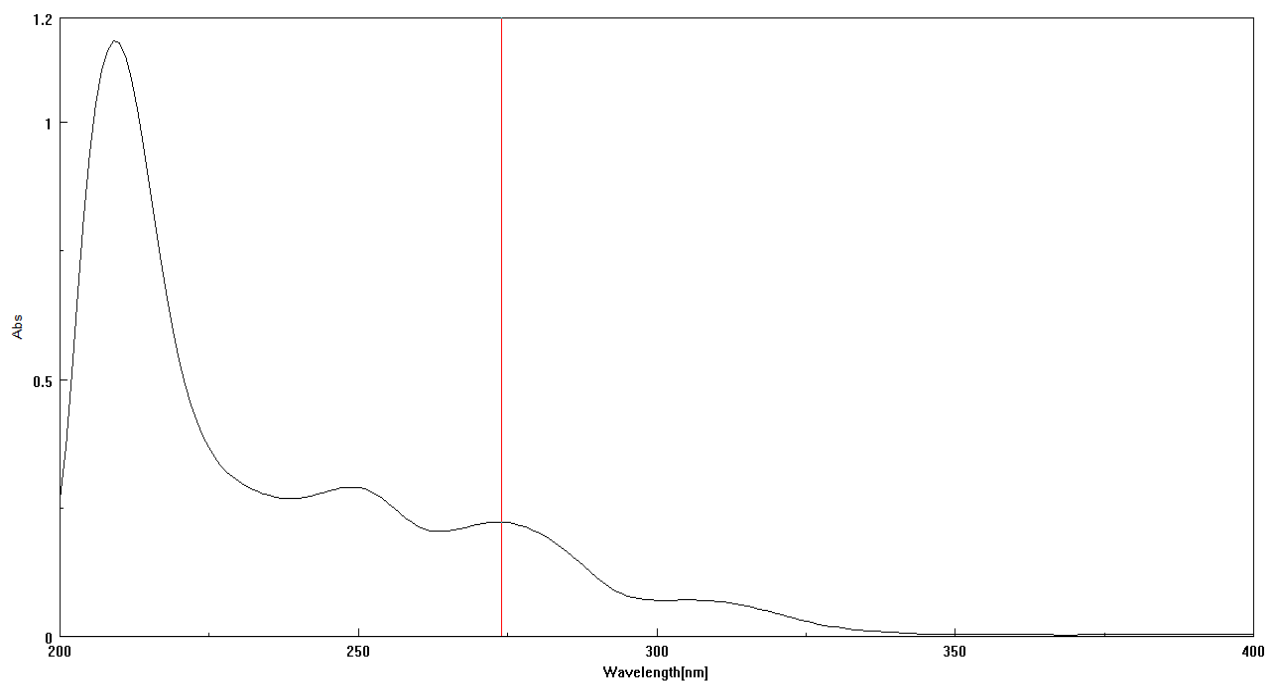
Figure 2: UV spectra of acebrophylline in ethanol.
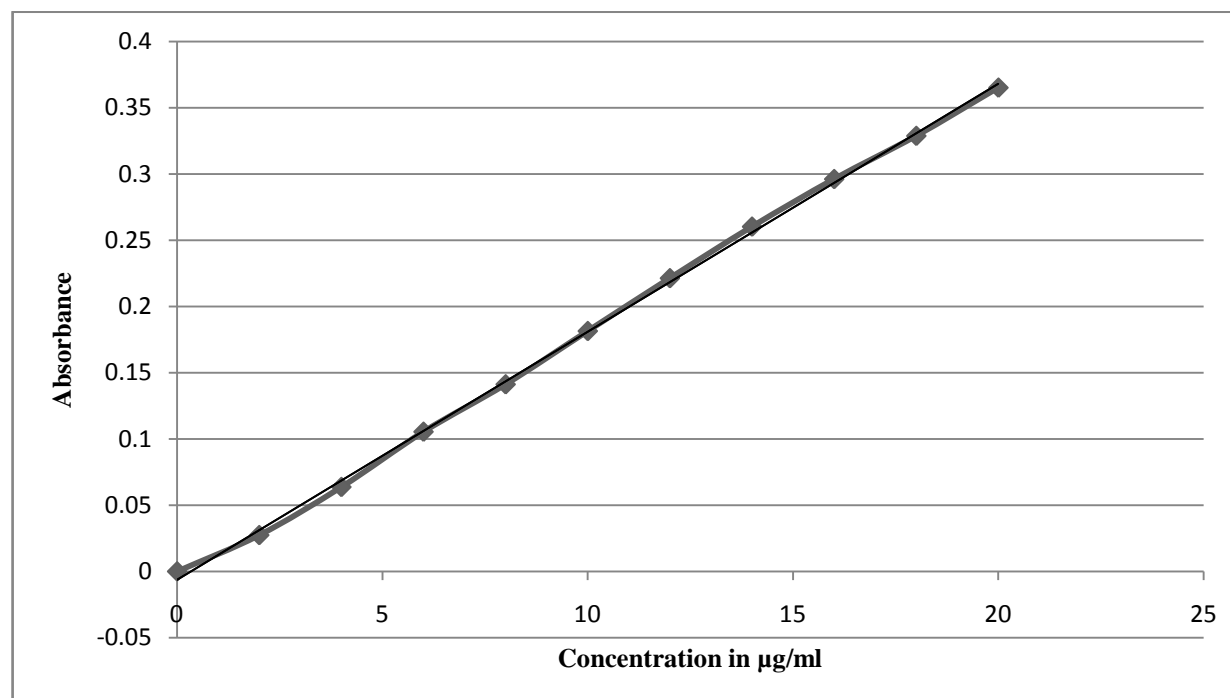
Figure 3: Calibration curve for Acebrophylline.

Table 1: Optical parameter.

Sr. No.	Parameter	Observation
1	Absorption maxima	274nm
2	Linearity range	2 to 20 µg/ml
3	Standard regression equation	Y = 0.0187X - 0.0062
4	Correlation coefficient (r^2)	0.9994
5	LOD in µg/ml	0.14117
6	LOQ in µg/ml	0.42780

Table 2: Recovery study of Acebrophylline (n=3).

Drug	Percentage addition	Amount added	Amount Recover	Recovery %	Average recovery
Acebrophylline	80	4	9.6396	101.790	100.1458
	100	5	9.3833	99.014	
	120	6	10.329	99.633	

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