Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Der Pharmacia Lettre, 2011, 3(3): 62-68
(http://scholarsresearchlibrary.com/archive.html)

ISSN 0975-5071
USA CODEN: DPLEB4

Spectrophotometric method for simultaneous estimation of Gatifloxacin and Ambroxol Hydrochloride in tablet dosage form

B.Prathap*, G.Nagarajan[1], A.Dinakar, G.Srinivasa Rao, Ranjit Singh B Rathor and Shahul Hussain

**Department of Pharmaceutical Analysis, Vagdevi College of Pharmacy and Research center, Nellore, A. P.*
[1]Department of Pharmaceutical Chemistry, PRIST University, Chennai

---

ABSTRACT

A simple, fast and precise simultaneous estimation method using Vierodt's method has been developed for the simultaneous determination of gatifloxacin and ambroxol hydrochloride in pure and combined tablet dosage form. Shimadzu UV-1700 instrument was used and λmax of gatifloxacin and ambroxol hydrochloride was found to be 289 and 246nm, respectively using methanol and 0.1M sodium hydroxide in the ratio of 8:2. Gatifloxacin and ambroxol hydrochloride were found to be linear in the concentration of 2-10µg/ml and 5-15µg/ml, respectively at their wavelengths. Amount found for gatifloxacin and ambroxol hydrochloride was found to be 397.61 and 73.91mg/tablet, respectively. Percentage recovery range was found to be within 99.45-100.3% for gatifloxacin and 99.73-103.42% for ambroxol hydrochloride. The percentage RSD was found to be lower than 2% proves that the method is precise. The application of simultaneous equation method by UV spectroscopy can be employed for the estimation of gatifloxacin and ambroxol hydrochloride for routine analysis for a combination containing these two components in pure and tablet dosage forms.

Key words: Gatifloxacin, Ambroxol hydrochloride, simultaneous equation method.

INTRODUCTION

Chemically gatifloxacin (GA) is 1-cyclopropyl-6-fluoro-8-methoxy-7-(3-methyl-1-piperazinyl)-4-oxo-3-quinolinecarboxylic acid sesquihydrate. GA is the synthetic broad spectrum 8-methoxy fluoro quinolone antibacterial drug, used in the treatment of community-acquired pneumonia, acute bacterial sinusitis, acute bacterial exacerbation of chronic bronchitis and complicated and uncomplicated urinary tract infections. Its acts intravenously by inhibiting topoisomerase II (DNA gyrase) or topoisomerase IV [2, 3 ,4]. Its empirical formula for GA is $C_{19}H_{22}FN_3O4$. Its

structure is given in Fig.1.

Ambroxol hydrochloride (AM) is a mucolytic agent. Chemically AM is Trans -4-[(2-amino-3, 5-dibromobenzyl) amino] cyclohexanol hydrochloride. The empirical formula for AM is $C_{13}H_{18}Br_2N_2O.HCl$ [1, 3 ,4]. Its structure is given in Fig.2.

A literature survey reveals the analytical methods like UV, HPLC and HPTLC and available for the determination of those drugs individually and other combinations in pharmaceutical and biological preparations [5-24]. However, there is no work has been reported for the estimation GA and AM simultaneously. In the present investigation an attempt was made to develop a simple and economical spectrophotometric method with greater precision, accuracy and sensitivity for the simultaneous estimation of GA and AM in pure and tablet dosage forms.

Fig. 1 Structure of gatifloxacin sesquihydrate

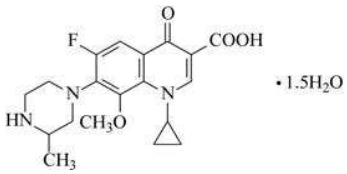
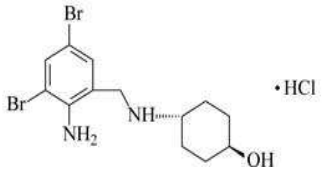


Fig. 2 Structure of ambroxol hydrochloride



MATERIALS AND METHODS

Experimental

Methanol and sodium hydroxide pellets were of analytical grade. Spectral absorbance measurements were made on shimadzu UV-1700 with 10mm matched quartz cell. The commercially available tablets were procured from the local market.

Solvent solution

The solvent solution used for this study was methanol: 0.1M sodium hydroxide which was prepared in the ratio of 8:2

Preparation of standard stock

Standard stock solution of gatifloxacin and ambroxol HCl were prepared separately by dissolving 400mg of gatifloxacin standard individually in solvent solution and made up to 100ml volume with solvent solution dissolving about 75mg of ambroxol HCl standard individually in solvent solution and made up to 100ml volume with solvent solution.

Working standard solution

From the above stock solution 1and 1.5ml of gatifloxacin and ambroxol HCl were transferred to 100ml volumetric flasks which were then made up to volume with mobile phase to give final concentration of 40 and 60µg/ml of gatifloxacin and 7.5 and 15 µg/ml of ambroxol HCl. The optical characteristics of gatifloxacin and ambroxol HCl were shown in Table-1.

Table-1 Optical characteristics and statistical data of the regression equation

Parameters	Gatifloxacin	Ambroxol HCl
Absorption maximum (nm)	289	246
Beer's law limit (µg/ml)	2-10	5-15
Correlation coefficient	0.9907	0.9896
Regression equation	0.0549X+0.025	0.0383X-0.0572
Intercept (a)	0.025	-0.0572
Slope (b)	0.0549	0.0383

Linearity and calibration

The stock solution of gatifloxacin was suitably diluted with solvent solution to provide varying concentrations of 2,4,6,8 and 10µg/ml which were scanned at its respective absorption maxima and the absorbance were plotted against concentration.

The stock solution of ambroxol HCl was suitably diluted with solvent solution to provide varying concentration of 5,7.5,10,12.5 and 15 µg/ml, which were scanned at its respective absorption maxima and the absorbances were plotted against concentration. From the graph it was found that the Beer,s law limit lies between 2-10µg/ml for gatifloxacin and 5-15µg/ml for ambroxol HCl. The regression analysis was carried out for calibration graph to find out correlation coefficient (r), intercept and slope of the regression line which were estimates the degree of linearity. Correlation coefficient was found out to be 0.9907 for gatifloxacin and 0.9896 for ambroxol HCl respectively. The overlain spectra of gatifloxacin and ambroxol HCl were depicted in Figs. 1 and 2.

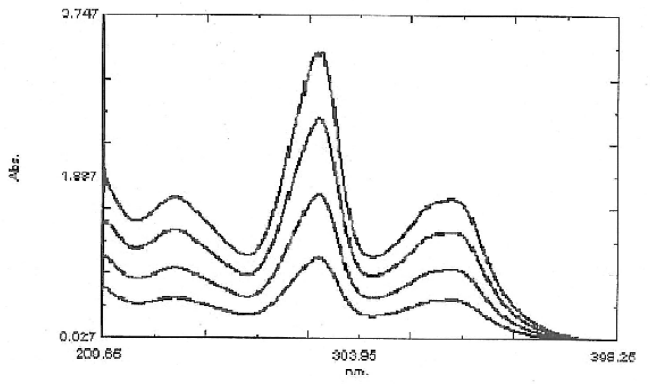


Fig1. Overlay spectra of gatifloxacin by UV method

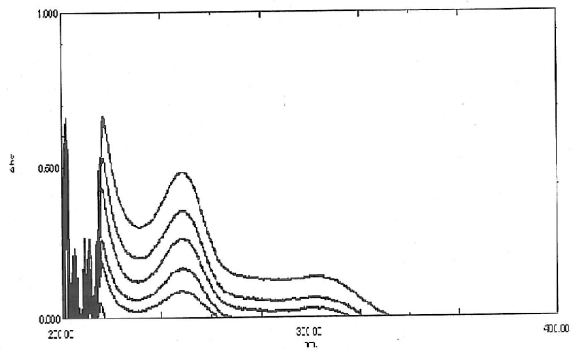


Fig 2. Overlay spectra of ambroxol HCl by UV method

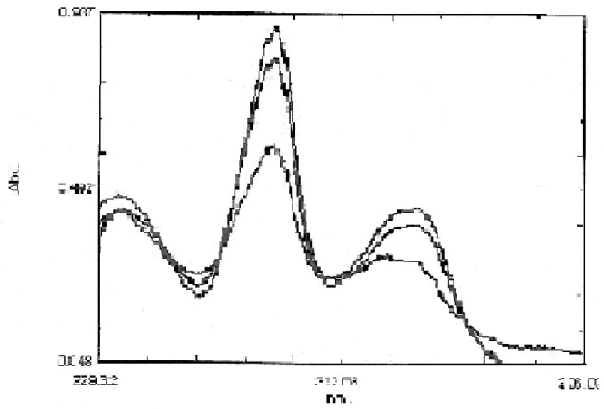


Fig 3. Overlay spectra of sample formulation by UV method

Simultaneous equation method [25]

Preparation of sample solution

Twenty tablets were weighed accurately and grounded to fine powder. An accurately weighed quantity of powder equivalent to 400 mg of gatifloxacin and transferred to 100 mL volumetric flask. The contents in the flask was dissolved with minimum amount of solvent solution, sonicated for 0.5 h and then diluted to 100 mL with solvent solution. The resultant was filtered with Whatmann filter No. 4. The filtrate was further diluted to made concentrations of 20, 40 and 60 µg/mL of gatifloxacin and 3.5, 7.5, 15µg/mL of ambroxol HCl, respectively. These dilutions were scanned over the range 200-400 nm.

Assay

Mixed standards and sample solutions were scanned over the range of 200-400 nm in the spectral mode. The concentration of each component was obtained by analysis of the spectral data of sample solution with reference to that of three mixed standard dilutions in the spectral mode of analysis.

The concentration of each component was obtained by analysis of the spectral data of sample solution using the Cramer's rule. Amount of drug present in sample was calculated and given in Table-2. Statistical analysis data's were presented in Table-3 and the overlain spectra of sample formulation was shown in Fig. 3.

Table-2 Assay Of Sample Formulation By Simultaneous Equation Method

Sample	Lable claim (mg/tab)	Amount fount (mg)	Lable claim (%)
Gatifloxacin	400	397.61	99.4
Ambroxol HCl	75	73.91	98.6

Table-3 Statistical Validation

Drug	Lable claim (mg/tab)	% amount estimated	% R.S.D
Gatifloxacin	400	397.61	1.74
Ambroxol HCl	75	73.91	2.02

Method validation

As per ICH guidelines the method is validated and following parameters were evaluated. Accuracy of the method was checked by recovery studies. Precision of the method was studied by inter-day and intra-day analysis of multiple samplings of homogenous sample and expressed as % RSD.

Recovery studies

Accuracy, specificity of the proposed method was satisfied by conducting recovery studies. Recovery studies was carried out by mixing a known quantity of standard drug in three levels to pre analyzed sample solution and the contents were reanalyzed by the proposed method. The data were given in Table-4.

Table-4 Recovery Data

Drug	Amount added (µg)	Amount recovered* (µg)	% Recovery*	Average Recovery (%)	% RSD
Gatifloxacin	20	19.89	99.89	99.73	0.318
	40	40.12	100.3		
	60	59.72	99.53		
Ambroxol HCl	3.5	3.62	103.42	101.32	0.535
	7.5	7.48	99.73		
	15	15.12	100.8		

**Mean of three determinations at each level.*

Table-5 Repeatability Studies

Drug	Amount taken (µg/ml)	Inter day		Intra day	
		Amount found* (µg/ml)	% RSD	Amount found* (µg/ml)	% RSD
Gatifloxacin	20	19.73	0.535	19.87	0.473
	40	40.23	0.238	40.13	0.198
	60	59.89	0.183	59.88	0.157
Ambroxol HCl	3.5	3.472	0.307	3.489	0.199
	7.5	7.508	0.067	7.511	0.047
	15	14.871	0.866	14.905	0.573

**Mean of three determinations; RSD-relative standard deviation.*

Repeatability Studies

Repeatability is given by inter-day and intra-day precision. Intra-day precision was determined

by analyzing, the three different concentration of drug for three times in the same day. Inter-day precision was determined by analyzing the three different concentration of the drug for three days in a week and the results were presented in Table-5.

RESULTS AND DISCUSSION

Simultaneous estimation of gatifloxacin and ambroxol HCl was carried out by simultaneous equation method.

Absorption maxima: The absorption maxima of the proposed drug were determined scanning in the entire UV region between 200-400 nm. Absorption maxima were found to be 289 and 246 nm respectively.

Beer's law concentration range linearity: The method was validated by linearity studies which were performed by plotting five different concentrations of standard solution against their respective absorbances. Gatifloxacin and ambroxol HCl were found to be linear in the concentration range of 2-10, 5-15 µg/mL, respectively.Linearity was expressed in terms of linear regression equationYgatifloxacin = 0.0549x+0.025; YAmbroxol HCl = 0.0383x - 0.0572 and correlation co-coefficient values were found to be 0.9907 and 0.9896 for gatifloxacin and ambroxol HCl respectively.

Assay: The quantitative estimation was carried out on marketed tablets (Ecogat A) and the amount was found to be 397.61 and 73.91 mg/tablet respectively. The results obtained from the quantitative work were subjected to statistical analysis. The percentage purity values near to 100 % w/w and percentage RSD values less than 2 % values shows that the method is accurate.

Accuracy: The proposed method was further validated by recovery analysis performed by adding known concentrations of standard drugs to preanalyzed sample solution at three different levels. Percentage recovery range was found to be within 99.4-100.3 % for Gatifloxacin and 99.7-103.4 % for Ambroxol HCl.

Repeatability studies: Repeatability is given by inter-day and intra-day precision. Intra-day precision was determined by analyzing, the three different concentration of drug for three times in the same day. Inter-day precision was determined by analyzing the three different concentration of the drug for three days in a week were determined. The percentage RSD was found to be lower than 2 % proves that the method is precise.

CONCLUSION

The application of simultaneous equation method by UV spectroscopy can be employed for the estimation of gatifloxacin and ambroxol HCl for routine analysis for a combination containing these two components in tablet dosage forms.

REFERENCES

[1] British Pharmacopoeia, Vol. 1, **2003**, 104-105.

[2] United States Pharmacopoeia-25 (NF-20), pp. 1064.
[3] Martindale, the Complete Drug Reference, edn. 32, pp. 1574.2.
[4] The Merck Index, Merck and Co., edn. 13, p. 2001, 4391 and 382-383.
[5] Roosewelt C, Muthuprasanna P, Harikrishnan N, Srinivasa Rao and Gunasekaran V.*J. Pharm. Res.* **2006**, 5 (2), 51-52.
[6] Lakshmi sivasubramanian and muthukumaran A. *Ind. J.Pharm.Sci.* **2005**, 367-369.
[7] Jacob Jane, Subrahmanyam S and Sathyanarayana D. *Asian. J.Chem.* **2006**, 18(4), 3210-3212.
[8] Gandhimathi M, Manjuladevi A S, Ravi T K, Majeed .J and Francis D. *Ind. Drugs.* **2006**, 43(1), 31-34.
[9] Kuchekar SB, Shinde SG, Naikawadi I T, Todkar J and Kharade S V. *Ind.J.Pharm.Sci*, **2003**, 193-195.
[10] Anju goyal and Singh L. *Asian. J. Chem.* **2005**, 17(4), 2836- 2838.
[11] Saleh .A.I dgither, Syed Naseeruddin Alvi and Muhammad .M Hammami. *J.Pharm.Biomed.Anal.* **2006**, 41(1), 251-255.
[12] Brian .R Overholser, Michael .B Kays and Kevin .M Sowinski. *J. Chrom B.* **2003**, 798(1), 167-173.
[13] Sanjay .K Motwani, Roop .K Khar, Farhan .J Ahmad, Shruti Chopra, Kohil K, Sushma Talegaonkar and Zeenat Iqbal. *An.Chim.Acta*, **2006**, 576(2), 253-260.
[14] Hairui Liang, Michael .B Kays, Kevin and Sowinski. *J.Chrom B.* **2002**,772(1),53-63.
[15] Grzegorz Bazylak and Luc J Nagels. *J.Pharm.Biomed. Ana.* **2003**, 32 (4-5), 887-903.
[16] Maarit Heinanen and Coral Barbas. *J.Pharm.Biomed. Anal.* **2001**, 24(5-6), 1005-1010.
[17] Bhanubhai .N Suhagia, Shailesh .A, Inhwarsinh S, Haresh .M Patel, Dinesh .R Shan and Bhavin .P Marolia. *Anal.Sci.* **2006**, 22, 743-745.
[18] Sowmiya G, Gandhi M, Ravi T K and Siveesaa K R. *Ind.J.Pharm.Sci.* **2007**, 69(2), 301-302.
[19] Shan S A, Rathod I S, Suhagia B N and Baldaniya M. *Ind.J.Pharm.Sci.* **2004**, 66(3), 306-308.
[20] Prathap B, Nagarajan G, Roosewelt C, Gopal V. *Der Pharmacia Lettre***, 2010,** 2(3): 163-167.
[21] Boopathy D, Bibin Mathew, Prakash M, Sureshkumar S and Perumal P. *Der Pharmacia Lettre*, **2010**, 2(2): 346-349.
[22] Mothilal M, Pushpalatha R, Damodharan N, Manimaran V, Lakshmi K S. *Der Pharmacia Lettre*, **2010,** 2(3): 286-296.
[23] Swati C. Jagdale, Santosh S. Padekar, Ajay S. Bhadoriya, Swapnil A. Ghorpade, Bhanudas S. Kuchekar. *Der Pharmacia Lettre*, **2010**, 2(4): 217-226.
[24] A.H. Beckett, In Practical Pharmaceutical Chemistry, CBS Publishers and Distributors, New Delhi, edn. 4, **2001**, 157-167, 276-296.