

## Spectrophotometric and HPLC methods for simultaneous estimation of cefadroxil and ambroxol from tablets

ESTHER ROSET SHOBANA<sup>1</sup> and LAKSHMI SIVASUBRAMANIAN<sup>2</sup>

<sup>1</sup>Department of Pharmaceutical Chemistry, Vellore Institute of Technology,  
Vellore - 632 014 (India)

<sup>2</sup>Department of Pharmaceutical analysis, SRM college of Pharmacy,  
SRM University, Kattankulathur - 603 203 (India)

(Received: July 18, 2008; Accepted: August 19, 2008)

### ABSTRACT

A simple, accurate, economical and reproducible UV spectrophotometric and a HPLC method for simultaneous estimation of two-component drug mixture of cefadroxil and ambroxol in combined tablet dosage form have been developed. The first developed method employs multiwavelength spectroscopy using six mixed standards and 230.0 nm and 245.0 nm as two wavelengths for estimation. Linearity was observed in concentration range of 10-60 µg/ml of cefadroxil and 1.2-7.2 µg/ml of ambroxol. Developed HPLC method is reverse-phase chromatographic method using Hypersil C<sub>18</sub> column and acetonitrile:phosphate buffer (pH 5.0) in the ratio of 10:90 v/v as mobile phase. For HPLC method, linearity was observed in concentration range of 100 –500 µg/ml of cefadroxil and 12.5-62.5 µg/ml of ambroxol. Results of analysis were validated statistically and by recovery studies.

**Key words:**HPLC, Cefadroxil, Ambroxol tablets.

### INTRODUCTION

Cefadroxil, chemically 7-[(R)-2-amino-2-(4-hydroxy phenyl) acetamido]-3-methyl-3-cephem-4-carboxylic acid monohydrate is a first generation cephalosporin used in the treatment of respiratory tract and urinary tract infections. Cefadroxil is listed in Merck Index<sup>1</sup>. Ambroxol, chemically 4{[2-amino-(3,5-dibromo phenyl) methyl] amino}- Cyclohexanol is a mucolytic used in the treatment of acute and chronic respiratory tract disorders<sup>2,3</sup>. Literature survey indicated spectrophotometric<sup>4-6</sup>, HPLC<sup>7-11</sup> have been developed for the estimation of cefadroxil and ambroxol, individually and in combination with other drugs. But no method has been established so far, for the simultaneous estimation of these drugs in combined dosage form. The present investigation is an attempt to develop highly sensitive, precise and rapid analytical methods for

the simultaneous estimation of cefadroxil and ambroxol from tablet formulations.

### MATERIAL AND METHODS

Standard bulk drug samples of cefadroxil and ambroxol were provided by Madras Pharmaceuticals, Chennai. Tablets of combined dosage form were procured from the local market. All other reagents used were of analytical grade for spectrophotometric method and of HPLC grade for HPLC method. Shimadzu UV/Vis Spectrophotometer (model 1601) with 1 cm matched quartz cell was used for spectrophotometric method. Spectra were recorded using specific program of apparatus, having specifications as follows: spectral bandwidth 3 nm, wavelength accuracy ± 0.5 nm, wavelength readability 0.1 nm increments. For HPLC method,

Shimadzu delivery module LC-10AD with UV SPD-10A detector was used.

#### Multiwavelength spectroscopy

Using the overlain spectra of cefadroxil and ambroxol in methanol, the wavelength maxima of both drugs, i.e., 230.0 and 245.0 nm, were selected as two sampling wavelengths for this method. Six mixed standards of two drugs in methanol were prepared so as to contain 10-60 µg/ml of roxithromycin and 1.2-7.2 µg/ml of ambroxol. All mixed standard solutions were scanned over the range of 400 to 200 nm in multicomponent mode of spectrophotometer using 230.0 nm and 245.0 nm as two sampling wavelengths. The spectral data from these scan were used to determine the concentration of two drugs in the sample solution.

#### Analysis of commercial formulation

Twenty tablets were accurately weighed and average weight per tablet was determined. Tablets were ground to fine powder and weighed tablet powder equivalent to 250 mg of cefadroxil was transferred to 100 ml volumetric flask.

The powder was dissolved in methanol by intermittent shaking and the volume was made upto the mark with methanol. The solution was then filtered through Whatman filter paper no:1. Aliquot of this solution was diluted to get a final concentration 30 µg/ml and 3.6 µg/ml of cefadroxil and ambroxol respectively. The sample solution was scanned over the range of 400 to 200 nm in multicomponent mode and concentration of each component was estimated by analysis of spectral data of sample solution with respect to that of mixed standards by the instrument. Results of analysis are reported in Table 1.

#### High Performance liquid chromatographic method

HPLC method was developed using Hypersil C<sub>18</sub> ODS (5µ) 250x4.6 mm column. Mobile phase selected for this method contained 90 parts of phosphate buffer (pH 5.0) and 10 parts of acetonitrile that was filtered through 0.45 µ membrane filter. Flow rate employed was 1.5 ml/min. Detection of eluent was carried out at 230 nm.

**Table 1: Results of analysis of commercial formulation**

Method	Label Claim (mg/tablet)		% Label claim		Standard Deviation	
	CE	AM	CE	AM	CE	AM
Method 1 (UV)	250	30	100.18	100.10	0.5252	0.667
Method 2 (HPLC)	250	30	100.06	100.05	1.147	0.835

CE – Cefadroxil, AM – Ambroxol and \* - Average of five determinations

**Table 2: Results of Recovery Studies**

Method	Concentration added (mcg/ml)		% Concentration recovered	
	CE	AM	CE	AM
Method 1(UV)	10	1.2	101.52	99.72
	20	2.4	99.67	100.06
	30	3.6	100.08	100.85
Method 2(HPLC)	100	12.5	99.1	102.70
	200	25	101.2	101.50
	300	37.5	99.87	101.70

CE – Cefadroxil, AM – Ambroxol and \* Average of Three determinations

### Standard stock solution

Standard stock solutions of pure drugs was made separately in mobile phase containing 1000 µg/ml of cefadroxil and 125 µg/ml ambroxol and filtered through a 0.45 micron membrane filter.

### Preparation of calibration curve

For preparation of the drug solutions for the calibration curves in a series of 10 ml volumetric flasks 1, 2, 3, 4, 5 and 6 ml of the pure drug standard stock solution containing 1000 µg/ml of cefadroxil and 125 µg/ml of ambroxol were transferred. The volume in each flask was made up to the mark with the mobile phase. About 20 µl of each solution was injected and chromatograms were recorded. Mean retention time for cefadroxil was found to be 2.51 min and for ambroxol 7.43 min with the resolution factor of 9.1. The peak areas of cefadroxil and ambroxol were measured and respective calibration curves were plotted against concentration of drug and peak area of drug. The tailing factor and HETP was found to be 1.32 and 6433 for cefadroxil and 1.47 and 3901 for ambroxol respectively.

### Procedure for analysis of formulations

Twenty tablets of the formulation were weighed and average weight per tablet was calculated. Twenty tablets were crushed and ground to a fine powder. Powder equivalent to 100 mg of cefadroxil was weighed and transferred to a 100 ml volumetric flask containing 50 ml mobile phase. The

powder mixture was dissolved in the mobile phase with the aid of ultrasonication. The solution was filtered through whatman filter paper no.41 into another 100 ml volumetric flask. The filter paper was washed with mobile phase and washings were added to filtrate. Volume of filtrate was made up to the mark with the mobile phase. To another 10 ml volumetric flask, 1ml of this solution was transferred and the volume was made up to the mark with the mobile phase. This solution was filtered through a 0.45µ membrane filter. After setting the chromatographic conditions and stabilizing the instrument to obtain a steady baseline, the tablet sample solution was loaded in the 20µl fixed sample loop of the injection port. The solution was injected and a chromatogram was recorded. The injections were repeated five times and the peak areas were recorded. A representative chromatogram has been given in Fig.1. The peak areas of each of the drugs were calculated and the amount of each drug present per tablet was estimated from the calibration curves. The results of analysis are presented in Table 1.

### Recovery studies

To study the accuracy, reproducibility and precision of the above methods, recovery studies were carried out by addition of standard drug solution to pre-analyzed sample at three different levels. Results of recovery studies were found to be satisfactory and are reported in Table 2.

## REFERENCES

1. The Merck Index, 12<sup>th</sup> Edition, Merck Research Lab, Rahway, N.J, 1996, 1925.
2. Reynolds, J.E.F., Eds., In: Martindale; The Extra Pharmacopoeia, 30<sup>th</sup> Edn., The Pharmaceutical Press, London, 1993, 743. The Merck Index, 12<sup>th</sup> Edition, Merck Research Lab, Rahway, N.J, 8430 (1996).
3. The Merck Index, 12<sup>th</sup> Edition, Merck Research Lab, Rahway, N.J, 8430 (1996).
4. Yun Prince A, McMillin J and Welch L, *J.Chromatogr*, **712**: 145 (1998).
5. El-Gindy A, El Walily A and Bedair M, *J. Pharm. Biomed. Anal*, **23**: 341-52 (2000).
6. Gallo Martinez L, Sevillano- cabeza A, Campins Falco P and Bosch-Reig F.A, *Anal. Chim. Acta*, **370**: 115 (1998).
7. Gallo Martinez L, Campins Falco P and Sevillano- cabeza A, *J. Pharm. Biomed. Anal*, **29**: 405-13 (2002).
8. Indrayanto, G. and Handayani, R., *J. Pharm.*

- Biomed. Anal.*, **8**: 781 (1993). *Biomed. Anal.*, **24**: 1005 (2001).
9. Dincer, Z., Besan, H., and Goger, NG., *J. Pharm. Biomed. Anal.*, **32**: 887 (2003). 11. Joshi S., *J. Pharm. Biomed. Anal.*, **28**: 795 (2002).
10. Heinanen, M and Barbas, C., *J. Pharm.*