Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2017, 9(3):24-28

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Derivative Spectrophotometric Estimation of Amoxicillin Trihydrate and Carbocisteine by Third Order Derivative Spectroscopy Method in Combined Dosage Form

Rajan V Rele

Central Research Laboratory, D.G. Ruparel College, Matunga, Mumbai, India

---

ABSTRACT

In the proposed method UV spectrophotometric, third order derivative method was developed for validation of amoxicillin tri-hydrate and carbocisteine from pharmaceutical formulation. The ICH guidelines were used for validation of amoxicillin and carbocisteine. The derivative method involves the measurement of absorbance, hence wavelengths 236.2 nm and 210 nm were found to be suitable for the estimation of amoxicillin and carbocisteine respectively. The linearity was found in the concentration range of 10 to 100 µg /ml for amoxicillin and 1 to 10 µg /ml for carbocisteine respectively. For amoxicillin the percentage mean recovery was found to be 100.075 % and for carbocisteine was 100.031 % respectively. The linearity, accuracy and precision of method were validated by statistically. The less percentage (%) RSD values indicates both intra and inter day have high grade of precision.

Keywords: Third order derivative UV spectrophotometric estimation; Amoxicillin; Carbocisteine

---

INTRODUCTION

Amoxicillin tri-hydrate is semi- synthetic penicillin. It is described as 6 - (D - 4 hydroxy phenyl glycyl amino) penicillin acid tri-hydrate.. It is generally used as antibacterial. Amoxicillin tri-hydrate is official in USP [1], IP[2] and BP [3]. Carbocisteine is a mucolytic drug, described chemically as (2R)-2-amino-3-[(carboxy-methyl) sulphanyl] propanoic acid, which breaks down mucus in the body so that it can be more easily cleared from the body. Carbocisteine is official in British Pharmacopoeia [3] and European Pharmacopoeia [4]. In literature HPLC[5-7] Spectrophotometric [8-10] methods were reported for validation of combined dosage form. This proposed simple method can also be used for the routine analysis of this combination formulation. In the proposed work development, optimization and validation of the method are presented.

EXPERIMENTAL SECTION

Materials and methods

Instrument and reagents:

For spectral study a Shimadzu UV-spectrophotometer, model 1800 (Shimadzu, Japan) was used with spectral band width of 0.5 nm by using a pair of 10 mm quartz cells. The UV-Probe 2.42 software was used for all spectral measurements. The amoxicillin and carbocisteine, reference standards with certificate analysis were obtained from reputed firm.

Preparation of standard drug solution:

1. Solution A: 1000 µg /ml standard amoxicillin tri-hydrate in 0.1N hydrochloric acid.

2. Solution B: The solution A was further diluted to give 100 µg/ml
3. Solution C: 1000 µg /ml standard carbocisteine in 0.1N hydrochloric acid.
4. Solution D: The solution A was further diluted to give 100 µg/ml

Preparation of sample solution

The average weight of powder from each capsule was determined. From the powder blend, 250 µg /ml of amoxicillin and 150 µg /ml of carbocisteine sample solution was prepared and used for further analysis.

Selection of analytical wavelength

A 100 µg/ml solution of amoxicillin and carbocisteine were scanned in the wavelength range from 400 nm to 190 nm. The UV probe 2.42 software was used for conversion of zero order spectrum to third order derivative spectrum. From the spectrum, 236.2 nm. and 210 nm were selected for study of amoxicillin and carbocisteine respectively.

Preparation of linearity curves

The aliquots in the range of 10 – 100 µg/ ml of amoxicillin trihydrate and 1 -10 µg/ ml of carbocisteine were used for linearity study. Solutions were scanned in the spectrum mode and zero order spectra were converted to third order derivative spectra. The overlain third order spectrum of amoxicillin trihydrate and carbocisteine were represented in Figures 1(a), 1(b) respectively.

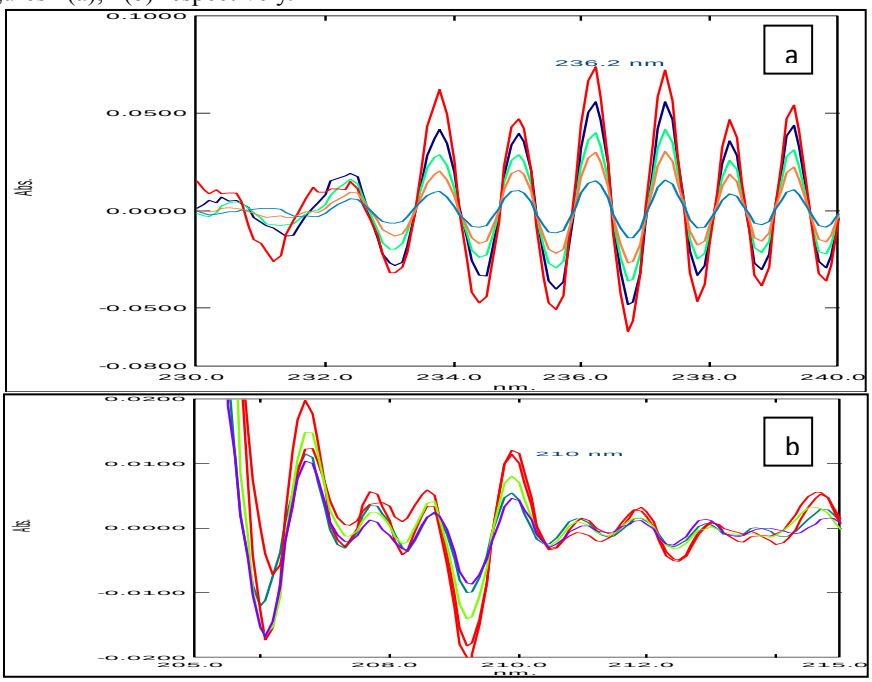

Figure 1: (a): overlain spectra of amoxicillin in the concentration range of 20 – 100 µg/ ml. (Third order derivative); b: overlain spectra o f carbocisteine in the concentration range of 2 – 10 µg/ ml. (Third order derivative)

The linearity curves were plotted of amplitude of third order derivative against concentrations (Figures 2 (a), 2(b)). Values of optical and regression of drugs analysis are given in table 1.

Table 1: Values of optical and regression of drugs

Terms	Carbocisteine	Amoxicillin trihydrate
Detection Wavelength (nm)	210	236.2
Beer Law Limits (µg/ml)	01-Oct	10-100
Correlation coefficient(r^2)	0.9973	0.9999
Regression equation (y=b+ac)		
Slope (a)	0.0013	0.0008
Intercept (b)	-0.0005	0.0002

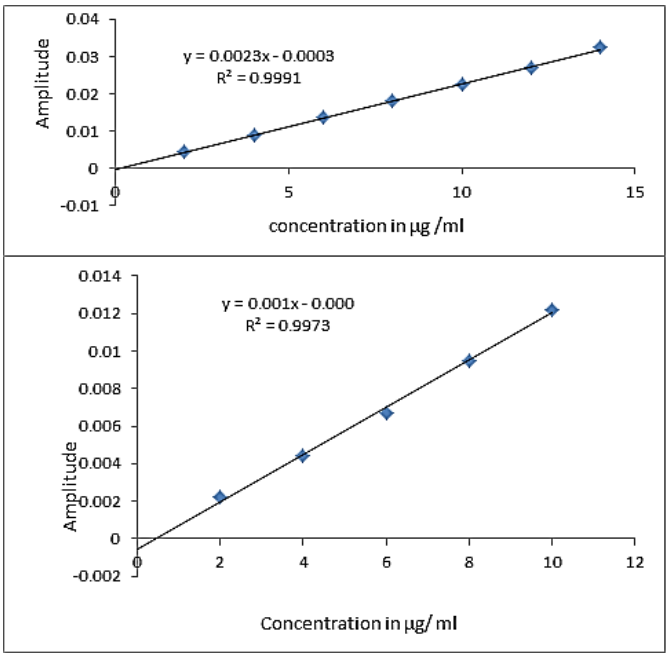

Figure 2 (a): Calibration curve of amoxicillin in the concentration range of 20-100 µg/ml; (b): Calibration curve of carbocisteine in the concentration range of 2-10 µg/ml

Estimation from capsules

The average weight of powder from each capsule was determined. From the powder blend, 250 µg /ml of amoxicillin and 150 µg /ml of carbocisteine sample solution was prepared and used for further analysis. The zero order spectra were converted to third order derivative spectra. Calculations were done as per the regression equations. The concentrations of amoxicillin and carbocisteine present in capsules were estimated by substituting the values of absorbance in equations.

a. For amoxicillin Y = 0.0008x + 0.0002
b. For carbocisteine Y = 0.0013x - 0.0005

Method validation

For validation ICH guidelines were used.

Accuracy, linearity and precision

For the determination of accuracy standard addition method at three different levels (80%, 100% and 120%) was car ried out. For amoxicillin and carbocisteine the Percentage recovery was found in the range of 99.88 % to 100.42 %. The linearity study was done by analyzing different aliquots of standard solutions of amoxicillin and carbocisteine a nd was found to be 10-100 µg/ml for amoxicillin and 10-100 µg/ml for carbocisteine. The precision study was perfo rmed by carrying out the analysis of powder blend from capsules.The 250 µg /ml of amoxicillin and 150 µg /ml of c arbocisteine sample solution was used in six replicates. The values of 0.1710 % for amoxicillin and 0.1959 % for car bocisteine were found for relative standard deviation were respectively, shown in Tables 2 and 3.

Table 2: Accuracy, statistical evaluation data

Stage	Initial amount present (µg/ml)		Amount of sample added (µg/ml)		Amount of drug found (µg/ml)		% Recovery		Mean % recovery	
	CARB	AMO	CARB	AMO	CARB	AMO	CARB	AMO	CARB	AMO
80%	15	25	12	20	26.964	44.919	99.87	99.82	99.88	99.915
	15	25	12	20	27.032	45.081	100.12	100.18		
	15	25	12	20	26.905	44.95	99.65	99.89		
100%	15	25	15	25	30.081	50.125	100.27	100.25	100.41	100.42
	15	25	15	25	30.321	50.085	101.07	100.17		
	15	25	15	25	29.952	50.06	100.65	100.12		
120%	15	25	18	30	32.94	54.791	99.82	99.62	100.07	99.89
	15	25	18	30	32.044	55.088	100.14	100.16		
	15	25	18	30	33.089	55.231	100.27	100. 42		

CARB = Carbocisteine AMOX = Amoxicillin tri-hydrate

Table 3: Statistical evaluation of the data subjected to method of precision

S. No.	Sample No.	% Assay	
		Carbocisteine	Amoxicillin Trihydrate
1	1	99.86	99.85
2	2	100.18	100.26
3	3	99.95	100.14
4	4	100.12	99.88
5	5	100.29	100.21
6	6	99.79	100.11
Mean % assay		100.031	100.075
% R.S.D.		0.1959	0.171

Intra-day precision was estimated by using the 250 µg /ml of amoxicillin and 150 µg /ml of carbocisteine sample sol ution in six replicates. Inter-day precision was estimated by using the 250 µg /ml of amoxicillin and 150 µg /ml of ca rbocisteine sample solution for three consecutive days (i.e. 1st, 3rd and 5th days). The statistical validation data for int ra and inter day precision is reported in table 4.

Table 4: Summary of validation parameter for intra-day and inter-day

S. No.	Study	Carbocisteine	Amoxicillin
1	Intra-day precision (N=3)amount found ± % R.S.D.	100.12%	100.24%
		0.1712	0.1633
2	Inter-day precision (N=3)amount found ± % R.S.D.	99.55%	99.45%
		0.1814	0.1548

The less values, in % RSD values of intra- day and inter-day indicates high degree of precision of the method.

RESULT AND DISCUSSION

The developed UV-spectrophotometric, third order derivative method for the assay of amoxicillin and carbocisteine in capsule formulation was convenient for the routine analysis of two drugs. The method is used to eliminate the spectral interference of one drug with other drug. The proposed method is reproducible. It is confirmed from validation data as subjected in tables 1 to 4. The Linearity was observed by linear regression equation method in different concentration. The correlation coefficient of these drugs was found to be less 1.00. It indicates good linearity.

CONCLUSION

The method does not require any ratio of third order derivatives. The amplitude of third order derivative can be directly used to assay of formulation. This method can be strongly recommended as an alternative to the existing methods. It can be easily and conveniently adopted for routine quality control analysis.

ACKNOWLEDGEMENT

Authors express sincere thanks to the principal of D.G. Ruparel College, Dr. Tushar Desai, for providing infrastructure facilities.

REFERENCES

[1] The United States Pharmacopeia. United States Pharmacopeia convention Inc, Rockville, **2008**, 30 (4), 1208.
[2] Indian Pharmacopeia, Controller of Publication, Delhi, **2007**, 1, II, III.
[3] British pharmacopoeia. Licensing division HMSO, Norwich, **2003**, 357.
[4] European Pharmacopoeia 5.0, **2005**, 2.
[5] A Nadiminti; A Gunda; KR Tupally; AP Lakshmi; K Jakka; AS Nagubandi. *J Pharm Res*. **2012**, 5(4),1889-1895.
[6] SK Sarode; ST Pise; P Dhore; DR Mundhada; S Bhaskaran. *BioMedRx*, **2013**, 1(4), 360-362.
[7] VA Patil; AC Vispute; SB Bari; DA Patil. *Trade Science Inc*. **2013**, 13(5).
[8] RV Rele. *Der Pharmacia Lettre*, **2014**, 6 (5), 1-7.
[9] RV Rele. *Asian J Res Chem*, **2014**, 7(4), 412-416.

[10] RV Rele. *Int J Chem Sci*, **2014**, 12(4), 1587-1595.