Research Article

Simultaneous Determination of Sulfamethaxazole and Trimethoprim by Using UV-Visible Spectrophotometer

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ABSTRACT

Sulfamethaxazole and trimethoprim are sulfonamide antibiotic drugs which are used for the treatment of the urinary tract, lungs (pneumonia), ears, and intestinal infections. In present work simple and effective method like simultaneous estimation method has been developed for the estimation of both the drugs in bulk and pharmaceutical dosage forms. The λmax of sulfamethaxazole and trimethoprim are determined as 270nm and 271nm with using the solvent as methanol. From the standard graph it was revealed that the beers limit for both drugs follow the concentration range of 2-10µg/ml and 10-50µg/ml with correlation coefficients of 0.9988 and 0.9989.The analysis work validated for precision and optical characteristics were found satisfactory.

Keywords: Simultaneous equation, Sulfamethaxazole, Trimethoprim, UV-spectrophotometer, λmax.

INTRODUCTION

Chemically, Sulfamethaxazole is N-(5-methyl-3-isoxazolyl) sulfanilamide and trimethoprim is 2,4-diamino-5-(3,4,5, trimethoxybenzyl)pyrimidine, Co-trimoxazole was claimed to be more effective than either of its components individually in treating bacterial infections[1-5] acute maxillary sinusitis and otitis media in children[6,7] gastro-intestinal infections manifested as shigellosis.[8] Literature survey reveals that there is no simple suitable spectrophotometric method for simultaneous estimation of sulfamethaxazole and trimethoprim in combined dosage form. The research work signifies a simple, precise, less expensive method for determination of both drugs in combination form.

The aim of the present work was to develop simple, precise, accurate, cost effective spectrophotometric method and validate as per ICH guideline.

MATERIALS AND METHODS

Spectrophotometer used was ELICO SL-210 (double beam UV–Visible spectrophotometer), Sulfamethaxazole and trimethoprim tablets are obtained from market. The pure samples of sulfamethaxazole and trimethoprim were obtained as a gift sample from VISTHA pharmaceutical pvt. Ltd, Nalgonda. All the chemicals used throughout the work are analytical grade.

Method Development

Preparation of standard stock solution

Standard stock solutions for sulfamethaxazole and trimethoprim were prepared by transferring accurately weighed 25mg of each into 25ml volumetric flask and the volumes were made up to 25ml using methanol as solvent to get 1000µg/ml.

Preparation of standard curve

From the standard stock solution fresh aliquots were pipette out and suitably diluted with methanol to get final concentration in the range of 10µg/ml separately. The solutions were scanned 200 to 400nm wavelength range and a sharp peak was obtained at 270 and 271nm. (figure1).

Preparation of standard graph

Different aliquots of standard solution of sulfamethaxazole ranging from 0.02-0.1 ml and 0.1-0.5 ml for trimethoprim were taken from 1000µg/ml were transferred in to series of 10 ml volumetric flasks separately. The volume in each flask was made up to 10ml with methanol the absorbances were measured at 270nm for sulfamethaxazole and 271nm for trimethoprim against methanol as blank. The obtained absorbances were plotted against the concentration to get the Standard graph. Calibration curve was plotted by taking absorbance on y-axis and concentration of solution on x-axis (figure2).

Overlain spectra of Sulfamethaxazole and Trimethoprim

Overlain spectrum has been drawn by choosing suitable solutions of both drug solutions and by using software spectra treats 3.11.01 Rel2.(Figure 3)

Estimation of Sulfamethaxazole and Trimethoprim in sample

Methodology[10,11]

Simultaneous equation method uses two selected wavelengths, one is λmax of Sulfamethaxazole and other is λmax of Trimethoprim. The stock solutions of both the drugs were further diluted separately with methanol to get a series of standard solutions of 2-10µg/ml for

Sulfamethaxazole and 10-50µg/ml for Trimethoprim. Concentrations in the sample were obtained by using these equations.

Cx =A1ay2-A2ay1/ax1ay2-ax2ay1.....eq-1

CY =A1ax2-A2ax1/ay1ax2-ay2ax1.....eq-2

A1 and A2 are absorbances of mixture at 270nm and 271nm.

Ax 1 and ax2 are absorptivitys of Sulfamethaxazole at λ1 and λ2 respectively.

Ay 1 and ay2are absorptivitys of Trimethoprim at λ1 and λ2 respectively.

Cx and Cy are concentration of sulfamethaxazole and trimethoprim respectively.

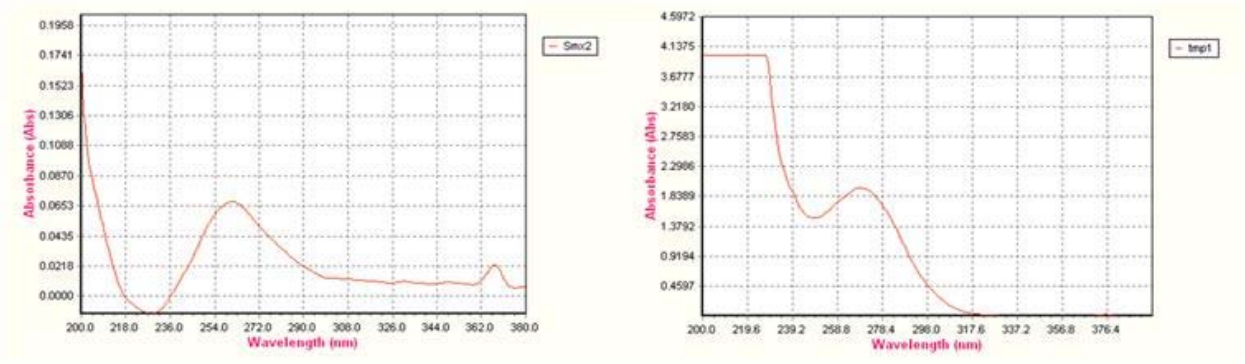

Figure 1: Spectra of sulfamethaxazole and trimethoprim in methanol

Table 1: Linearity results of sulfamethaxazole and trimethoprim in methanol

Sulfamethaxazole		Trimethoprim	
Concentration(µg/ml)	Absorbance	Concentration (µg/ml)	Absorbance
2 µg/ml	0.014	10 µg/ml	0.4523
4 µg/ml	0.028	20 µg/ml	0.895
6 µg/ml	0.0444	30 µg/ml	1.2656
8 µg/ml	0.0567	40 µg/ml	1.691
10 µg/ml	0.0719	50 µg/ml	2.0502

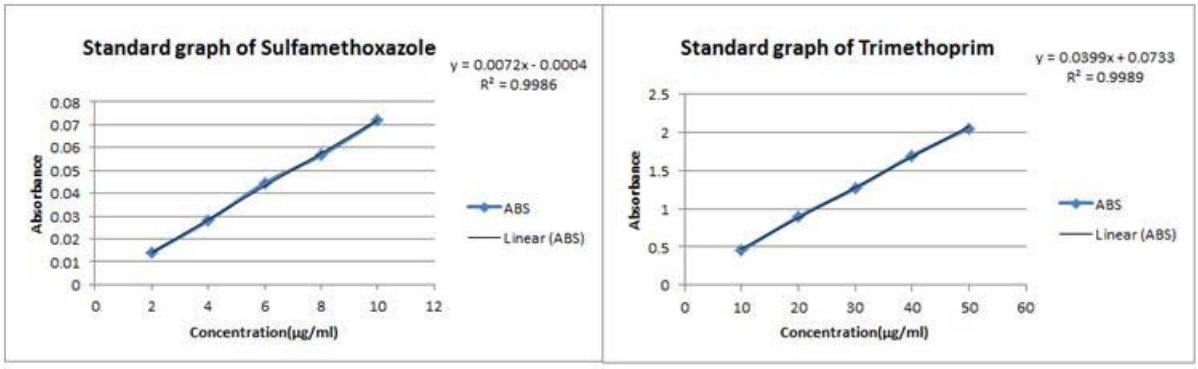

Figure 2: Calibration curve of sulfamethaxazole and trimethoprim

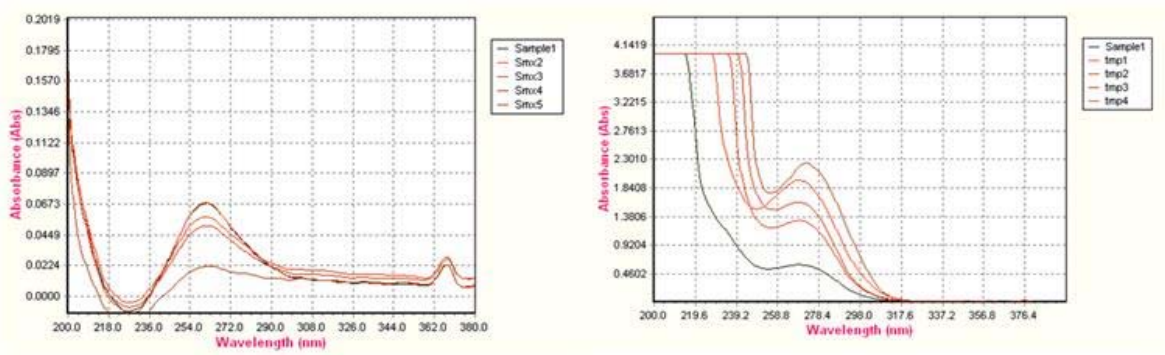

Figure 3: Overlain Spectra of sulfamethaxazole and trimethoprim in methanol

Results of Commercial Tablet Analysis

Table 2: Assay results of sulfamethaxazole and trimethoprim in methanol

Formulation	Labelledamnt.(mg)	Observed amnt. (mg)±S.D	% recovery	% RSD
Sample I	400+80	404.8±0.000767 79.1±0.000759	101.2 98.875	1.05244 1.0348
Sample II	400+80	403.01±0.00491 78.85±0.00487	100.752 98.57	1.09649 0.998

Validation Parameters

Precision

Table 3: Linearity results of sulfamethaxazole and trimethoprim in methanol

Sulfamethaxazole			Trimethoprim		
Concentration(µg/ml)	Absorbance		Concentration (µg/ml)	Absorbance	
10 µg/ml	0.0725	Mean = 0.07474	10 µg/ml	0.4469	Mean=0.43988
10 µg/ml	0.0739	SD = 0.000767	10 µg/ml	0.4498	SD=0.00491
10 µg/ml	0.0719	%RSD = 1.05244	10 µg/ml	0.4397	%RSD=1.09649
10 µg/ml	0.0733		10 µg/ml	0.4523	
10 µg/ml	0.0727		10 µg/ml	0.4502	

Optical characteristics

Table 4: Optical parameters

Parameters	Sulfamethaxazole	Trimethoprim
λmax	270nm	271nm
Beer's limit	2-10	10-50
Regression eqn.	Y=0.0072x-0.0004	Y=0.0399x+0.0733
Correlation coefficient	0.9986	0.9989
LOD	0.2767	0.3245
LOQ	0.7753	0.4169
Slope (a)	0.0072	0.0399
Intercept(b)	0.0004	0.0733

RESULTS AND DISCUSSION

In simultaneous determination method two wavelengths were used for analysis of drugs were 270nm and 271nm. The validation parameters were studied for both drugs at 270nm and 271nm. The % RSD was found to be1.05244 for sulfamethaxazole and 1.09649 for trimethoprim which are less than 2 shows the correctness of the result. The method wassuccessfully used to determine the amounts ofsulfamethaxazole and trimethoprimpresent in combined dosage form. By observing validation parameters the method was found to be simple, sensitive, accurate and precise. The % level claims in tablet were found to be100% to 102% for ciprofloxacin and 98% to 100% which is within the permissible limit. Hence the method can be employed for the routine analysis of these two drugs in their combined dosage form.

CONCLUSION

A proposed spectrophotometric method was found to be simple inexpensive and precise for determination of sulfamethaxazole and trimethoprim in their commercial formulation. The analysis of the mixture was done without any interference of excipients and additives. The simultaneous determination of the cited drugs in pure and tablet forms were done without any preliminary separation step so the present method is more economical and less time consuming compared to other chromatographic techniques.

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