Determination of Sulfamethoxazole in Pure and Pharmaceutical Samples by Using Direct Method for Calibration Curve of Normal Spectrum of UV-Spectrophotometry

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Articles Information	Abstract
	Normal spectra for sulfamethoxazole solutions were developed and used for the determination of sulfamethoxazole (SMX) antibiotic by using zero-crossing technique and simultaneously determining (SMX) at wavelength 259.00 nm. The correlation coefficient of the calibration curve for the normal spectrum was 0.9990. Linearity was maintained by using concentrations (0.990×10^{-4} M, 0.996×10^{-4} M, 0.999×10^{-4} M, 1.004×10^{-4} M, 1.005×10^{-4} M) and the percentage recovery of sulfamethoxazole samples were (99.00%, 99.60%, 99.90 %, 100.40 % and 100.50 %). A statistical analysis confirmed, a precision with accuracy for simultaneous fixing of (SMX). In addition, a British pharmacopoeia method was compared with the method used in this paper using F test.

1. Introduction

Sulfamethoxazole (SMZ) $C_{10}H_{11}N_3O_3S$, as shown in the Figure 1, is a crystalline and white powder, with molecular weight 253.279 g/mole, freely soluble in acetone, practically insoluble in the water, in ethanol sparingly soluble. As well as dissolves it in acids and in sodium hydroxide after diluted. Meanwhile also considered as a sulfonamide bacteriostatic antibiotic. In a synergistic combination, sulfamethoxazole is often used alongside trimethoprim in a ratio (5:1), in the co-trimoxazole [1]. Researchers have developed various analytical methods to determine sulfamethoxazole in biological samples and formulations, such as ion selective electrodes [2-4], chromatographic [5,6], and HPLC (High Performance Liquid Chromatography) [7-9] methods. UV spectrophotometer can be used to investigate the interaction of matter with light radiation in visible (400-800) range and the ultra-violet (200-400) [10,11]. UV radiation has enough energy to excite valence electrons in molecules or many atoms from their terrestrial state to higher energy levels. In such a process, excited electrons move from bond to anti-correlation orbits [12]. The typical applications of UV absorption spectroscopy comprise the determination of poly nuclear aromatic compounds such as vitamins, painting materials, and drugs [13]. In addition, several applications of visible spectrophotometric methods have been developed for the purpose of analysis of various colored compounds as well as, different colored metal complexes [13]. In this research, the determination of

Sulfamethoxazole was obtained in pharmaceutical samples using direct method for calibration curve of normal spectrum of UV-spectrophotometry at λ (259.00nm) for five samples of SMZ standard solution.
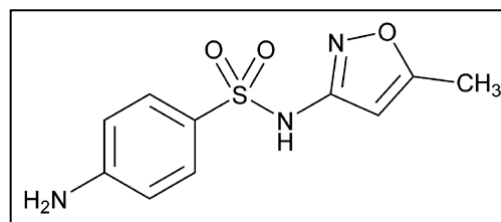

Figure 1. Structure formula of sulfamethoxazole.

2. Experimental Part

Instruments and equipment

The Shimadzu UV-1650 PC, UV-Visible, double-beam spectrophotometer (Japan) was interfaced via a data system of the Shimadzu UV probe software version 1.10, 1.00 cm quartz cells, (W. Germany) with Sartorius Handy of four-digit analytical Balance.

Reagents and solutions

The standard sulfamethoxazole, was a gift from the public company for Pharmaceutical and medical devices (Samara IRAQ-SDI). Trimol tablets were purchased each tablet (containing 400 mg sulfamethoxazole) from Bilim Pharmaceuticals (Made in UAE). The other reagents and solvents get from BDH.

3. Procedure
Preparation working standard solutions
1. The 20 % acetone solution was prepared by diluting 200 ml of acetone and supplementing it in a solution of 1000 ml with the distilled water.
2. Series of different solutions (0.990×10^{-4} M, 0.996×10^{-4} M, 0.999×10^{-4} M, 1.004×10^{-4} M, 1.005×10^{-4} M) were prepared from (1×10^{-3}) M sulfamethoxazole solution above.

4. Results and Discussion
Figure 2 shows the normal spectra for sulfamethoxazole solutions (0.990×10^{-4}, 0.996×10^{-4}, 0.999×10^{-4}, 1.004×10^{-4}, 1.005×10^{-4}) M, the absorption wavelength at 259.00 mm.
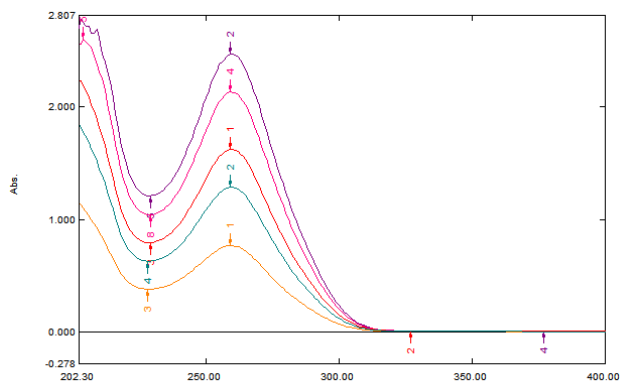

Figure 2. The normal spectra for SMZ solutions.

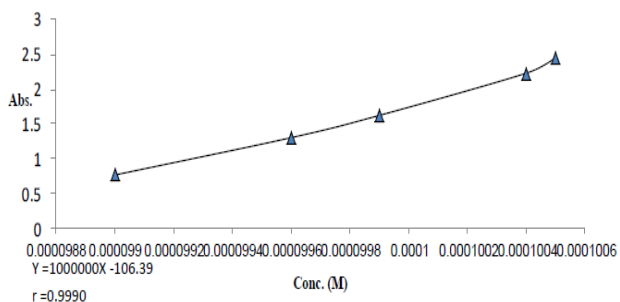The calibration curve was constructed for a wavelength (259.00) nm as shown in Figure 3.

Figure 3. Calibration curve of normal spectrum for SMZ at λ 259.00 nm.

Figure 3 shows the calibration curve of normal spectrum at the λ 259.00 nm, from a linear equation of the calibration curve ($Y = 1000000x - 106.39$) the concentration of sulfamethoxazole can be calculated. Table 1. Show the results of sulfamethoxazole.

Table 1. Calculation for five samples of 10-4of SMZ standard solution by using direct method for calibration curve of normal spectrum of UV-spectrophotometry at λ (259.00 nm).

Abs	Conc. (M)	δ_{n-1}*	X ± (ts/√N)	Re%	Er%	RSD%
0.764	0.990×10^{-4}			−1%	99.00%	
1.301	0.996×10^{-4}			−0.4%	99.60%	
1.621	0.999×10^{-4}	6.1400×10^{-7}	$0.998 \times 10^{-4} \pm 0.763 \times 10^{-6}$	−0.1%	99.90%	0.6152%
2.128	1.004×10^{-4}			0.4%	100.40%	
2.466	1.005×10^{-4}			0.5%	100.50%	

RSD: Relative standard deviation, Re%: The percentage recovery, Er: Relative error.

The calculated linear equation for normal spectrum, correlation coefficient and concentration are listed in Table 2.

Table 2. Calculation linear equations, correlation coefficient and the range of concentrations for the normal spectrum.

Method	Normal spectrum
nm (λ)	259.00
Concentration (M)	1.06×10^{-4}
Linear equation	y = 1000000x − 106.39
(r) Correlation Coefficient	0.9990

Comparison between ISE and normal spectrum methods

Table 3. The value of the F test between the ISE and normal spectra.

CU(M) from direct Method of ISE	δ_{n-1}*	CU(M) from direct method of normal spectrum	δ_{n-1}*	The (F) magnitude
0.993×10^{-4}		0.990×10^{-4}		
0.998×10^{-4}		0.996×10^{-4}		
0.992×10^{-4}	4.098×10^{-7}	0.999×10^{-4}	6.140×10^{-7}	2.244
1.002×10^{-4}		1.004×10^{-4}		
0.998×10^{-4}		1.005×10^{-4}		

δ_{n-1}*: standard deviation; n = 5, F = S12/S22, where S1 > S2, F Table = 6.39.

Table 3 shows the comparison between the (normal spectrum) and the (direct method) of ISE using the F-test using a 95% confidence level and this table show standard deviation (δ_{n-1}) for five measurements where F = S12/S22, where S1 > S2 and F Table equal 6.39 and from the calculations it found F Calculate is less than F Table, and thus, there is no difference in precision between the two methods. Sulfamethoxazole potentiometric determination in pharmaceutical preparations and the data obtained for pharmaceutical samples were listed in Table 4 [14].

Table 4. Analyses of SMZ in pharmaceutical samples.

Parameter	Direct method*	Standard addition method*	Multi SAM*	Titration method***
Conc. (Molar)	0.0001	0.0001	0.0001	0.0001
Found (Molar)	$0.998-10^{-4}$	0.998×10^{-4}	1.002×10^{-4}	0.999×10^{-4}
RSD*%	0.647	0.885	–	0.919
Re %	9 9.820	100. 980	100.200	99.95
Er %	−0.180	−0.360	0.200	−0.05
S	6.457×10^{-7}	8.820×10^{-7}	–	9.192×10^{-7}
X ± (ts/√N)	$0.998\times10^{-4}+0.779\times10^{-6}$	$0.998\times10^{-4} + 0.779\times10^{-6}$	–	$0.999\times10^{-4} +0.825\times10^{-5}$

RSD: Relative standard deviation, RSD***% for n = 2, t = 12.7, RSD**% for n = 3, t = 4.3, RSD*% for n = 5, t = 2.7, Re%: The percentage recovery, E_r: Relative error.

5. Conclusions

Analytical methods proved to be simple and fast. The results obtained from these methods are in good agreement, in terms of accuracy, with a direct method of selective ion electrode for analysis of the study using the F test at a 95% confidence interval compared with the normal spectrum.

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