Spectrophotometric Determination of Sulfamethoxazole Following Simple Diazotization and Coupling with Diphenylamine

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Received in: 7 May 2014,Accepted in: 22 june 2014

Abstract

Simple, sensitive, accurate and inexpensive spectrophotometric methods have been developed for the determination of sulfamethoxazole (SMZ) in pure and dosage forms. This method is based on diazotization of primary amine group of sulfamethoxazole with sodium nitrite and hydrochloric acid followed by coupling with diphenylamine in acidic medium to obtain a stable blue colored dye and show a maximum absorption (λ_{max}) at 530 nm. Different variables affecting the completion of reaction have been carefully optimized, following the classical univariate sequence and modified simplex method. Beer's law is obeyed in the concentration range of (0.5-12.0 µg.mL^{-1}) with molar absorptivity of 4.9617×10^4 L.mol^{-1}.cm^{-1}. The limit of detection was (0.108 µg.mL^{-1}), and Sandell's sensitivity value 5.1047 µg.cm^{-2}. The proposed method was successfully applied to the determination of sulfamethoxazole in pharmaceutical preparation.

Key words: Spectrophotometric, determination, Dosage forms, Sulfamethoxazole, Diazotization reaction, Coupling reaction, Diphenylamine.

Introduction

Sulfamethoxazole (SMZ) is a member of the sulfonamide family of antibacterial and chemically name is 4-Amino-N-(5-methyl-3-isoxazolyl)-benzene sulfonamide with molecular formula $C_{10}H_{11}N_3O_3S$ and molecular weight of (253.279 $g.mol^{-1}$), the basic structure of the drug is shown in Scheme (1). White and yellowish white colored, crystallized powder, its use has been limited by the development of resistance and it is now used mainly as a mixture with trimethoprim [1, 2, 3].

Mixture of sulfamethoxazole and trimethoprim which is known as co-trimoxazole is used to treat a wide variety of bacterial infections e.g.: middle ear infections, genito-urinary tract infections, respiratory-tract infections such as bronchitis, and enteric infections. Its main uses now are in Pneumocystis carinii pneumonia, toxoplasmosis, and nocardiosis. Gastrointestinal disturbances (mainly nausea and vomiting) and skin reactions are the most common adverse effects for this drug combination [4, 5]. Literature survey indicated that few analytical methods have been reported for analysis Sulfamethoxazole. They include some spectrophotometric method [6-10], HPLC [11- 18], flow injection analysis [19] and micellar electro kinetic capillary chromatography (MEKC) [20]. The aim of the present work is to provide an optimized spectrophotometric method using the univariate and multivariate simplex method. In the simplex method, three-interest factors concentration of sodium nitrite, diphenylamine, and hydrochloric acid were designated as independent variables and absorbance as response.

Experimental

Instruments

The absorption spectra were recorded, and all spectrophotometric measurements were carried out on (CECIL 7200) UV-Visible double beam spectrophotometer with 1 cm matched quartz cells.

Materials and Reagents

Pharmaceutical grade sulfamethoxazole powder received in pure form (99.99%) was provided as a gift from the State Company for Drug Industries and Medical Appliances Samara-Iraq (SDI). All chemicals and reagents used were of analytical grade.

Preparation of Solutions

*Sodium nitrite [0.2 % (m/v)]: prepared by dissolving 0.2 g of $NaNO_2$ in double distilled water and diluting to the mark in a 100 mL volumetric flask using distilled water**.**

*Sulfamic acid [1.0 % (m/v)]: prepared by dissolving 1.0 g of H_3NSO_3 in double distilled water and diluting to the mark in a 100 mL volumetric flask using distilled water.

*Diphenylamine (DPA) [0.5 % (m/v)]: prepared by dissolving 0.5 g of DPA in 25 mL methanol and diluting to the mark in a 100 mL volumetric flask with methanol.

*Hydrochloric acid [6 M]: prepared by diluting 52.52mL of concentrated HCl and diluted to 100 mL with distilled water.

*Hydrochloric acid [1M]: prepared by diluting 8.75 mL of concentrated HCl and diluted to 100 mL with distilled water.

*Hydrochloric acid [0.4M]: prepared by diluting 3.50 mL of concentrated HCl and diluted to 100 mL with distilled water.

Standard Drug Preparation (100 µg.ml^{-1})

The standard solution of SMZ was prepared by dissolving accurate weighted 0.01 g of pure drug in 10 mL of 0.4 M HCl and completed volume to the mark in volumetric flask 100 mL with distilled water. Working solutions were freshly prepared by subsequent dilutions.

Solution for the Analysis of Sulfamethoxazole in Pharmaceutical Preparations

i. In Tablets

The content of 10 tablets was grinded and mixed well. A certain amount of the fine powder was accurately weighted to give an equivalent to 800 mg for tablets and the mean value of the weight of one tablet was calculated .An amount of the powder equivalent to about 0.0126 gm. was accurately weighted, then about 10 mL of 0.4 M HCL was added, transferred into 100mL volumetric flask, and the solution was shaken swirled, left to stand for 5 mints and diluted to the mark in a volumetric flask 100 mL with distilled water to get 100 $\mu g.ml^{-1}$. The solution was filtered by using Whatman filter paper No.41 to avoid any suspended or un-dissolved material before use, and the first portion of the filtrate was rejected. Working solutions were freshly prepared by subsequent dilutions with distilled water, and analyzed by the recommended procedure.

ii. In Syrup

Each 5 mL of the syrup contains (200 mg of sulfamethoxazole with 40 mg of trimethoprim). An accurately measured volume (0.25 mL) was transferred into a 100mL volumetric flask, then added 10 mL of 0.4 M HCl swirled, left to stand for 5 mints and diluted to the mark with distilled water to get 100 $\mu g.mL^{-1}$ SMZ solutions. The solution was filtered by using Whatman filter paper No.41 to avoid any suspended or un-dissolved material before use, and the first portion of the filtrate was rejected. Working solutions were freshly prepared by subsequent dilutions with distilled water, and analyzed by the recommended procedure.

Determination of Sulfamethoxazole in Pharmaceutical Preparation by Standard Additions Method (SAM)

1- Preparation of SMZ stock solution 100 $\mu g.mL^{-1}$ according to the method of preparation previously mentioned.

2- Prepare solution of commercial pharmaceutical preparation (syrup or tablets) concentration of (100 $\mu g.mL^{-1}$); according to the method of preparation previously mentioned.

3- Three sets of unknown solution for three drug concentration levels namely; (10.0, 20.0 and 30.0 µg), each set was prepared by using seven 10.0 mL calibrated flasks by the addition of (0, 5.0, 10.0, 15.0, 20.0, 25.0 and 30.0 µg) from the standard stock solution of SMZ drug. After cooling in an ice bath, 1.0 mL of 0.2 % (m/v) sodium nitrite solution and 1.0 mL of 0.9 M HCl were added to each flask. The solution was shaken thoroughly; 1.0 mL of 0.30 % (m/v) sulfamic acid was added. The solutions were swirled and the resulting diazotized product was coupled with DPA by the addition of 1.0 mL of 1.0 % (m/v), this mixture was allowed to stand for 10.0 minutes. Each solution were making up to the mark with 6 M HCl. After mixing the solution well, the absorbance of blue colored dye was measured at 530.0 nm against the reagent blank. Figure (10, 11, 12 and 13) shows standard addition plot.

General Standard procedures

Two procedures were recommended for the determination of SMZ via the proposed methods. The first was carried out following the conditions obtained by univariate optimization, while the second based of those conditions obtained by chemometric multivariate simplex optimization**.**

Univariate Method

Aliquots of the standard solution (100 $\mu g.mL^{-1}$), containing (5-70) µg of sulfamethoxazole were transferred into a series of 10 mL volumetric flasks. After cooling in an ice bath, 1.0 mL of 0.02 % (m/v) sodium nitrite solution and 1.0 mL of 0.2 M HCl were added to each flask. The solution was shaken thoroughly; 1.0 mL of 0.3 % (m/v) sulfamic

acid was added. The solutions were swirled and the resulting diazotized product was coupled with DPA by the addition of 1.0 mL of 0.5 % (m/v) and allowed to stand for 10.0 minutes. The solutions were making up to the mark with 6 M HCl. After mixing the solution well, the absorbance of blue colored dye was measured at 530.0 nm against the reagent blank.

Simplex Method

Aliquots of the standard solution (100 µg.mL^{-1}), containing (5-120) µg of sulfamethoxazole were transferred into a series of 10 mL volumetric flasks. After cooling in an ice bath, 1.0 mL of 0.2 % (m/v) sodium nitrite solution and 1.00 mL of 0.90 M HCl were added to each flask. The solution was shaken thoroughly; 1.0 mL of 0.30 % (m/v) sulfamic acid was added. The solutions were swirled and the resulting diazotized product was coupled with DPA by the addition of 1.0 mL of 1 % (m/v) and allowed to stand for 10.0 minutes. The solutions were making up to the mark with 6 M HCl. After mixing the solution well, the absorbance of blue colored dye was measured at 530.0 nm against the reagent blank.

Results and Discussion

Absorption Spectra and Reaction Scheme

Primary aromatic amine upon treatment with nitrous acid in an ice bath, solution forms diazonium salt.

$$Ar\text{-}NH_2 + NaNO_2 + HCl \rightarrow Ar\text{-}N_2^+Cl^- + H_2O$$

Under proper conditions, diazonium salts react with certain aromatic compounds to yield products of general formula Ar-N=N-Ar`, called azo compounds and the reaction is called di azo coupling reaction [21].

$$Ar\text{-}N_2+ + Ar`H \rightarrow Ar\text{-}N=N\text{-}Ar`+ H^+$$

The proposed method involves coupling reaction of the diazotized sulfamethoxazole with diphenylamine in an acidic medium to yield an azo dye according to (Scheme 2). The former blue colored product shows a maximum absorption at 530 nm (Figure 1).

Optimization of Reaction Variables

Various parameters (viz effect of diazotization reaction time, effect of sodium nitrite concentration, effect of different acid, effect of acidity on diazotization, effect of sulfamic acid concentration, effect of diphenylamine concentration, effect of coupling reaction time, and effect of acidity) were first optimized, for the development of color dye, univariatly by systematic study of the effects of each parameters in the development of color product. The optimization steps were carried out by varying the parameters each one at a time and controlling all others fixed.

Effect of Diazotization Reaction Time

The optimum diazotization reaction time was determined at ~ 0-5 ºC by following the absorbance of the formed azo-dye for the period of (0-20) min. It was found that a colored product with maximum absorbance at 530.0 nm takes place instantaneous, after which no increase in absorbance values was obtained (Table 1), shows the results.

Effect of Sodium Nitrite Concentration

The effect of the concentration of $NaNO_2$ was studied by measuring the absorbance of the color products at 530.0 nm in the range of (0.01-0.30 % m/v) (Figure 2). It was found that 1.0 mL of 0.02 % solution sodium nitrite was needed for constant and maximum color intensity for azo dyes complex, low absorbance values were observed with higher concentrations.

Effect of Different Acid

Various acidic solutions (H_2SO_4, HCl, HNO_3, and CH_3COOH) with 1 M of concentration were tested for diazotization reaction. Among these, HCl was found to be the best by virtue of

highest absorbance values and stability considerations for diazotization; (Table 2) shows the results.

Effect of Acidity on Diazotization

The influence of hydrochloric acid concentration on the diazotization reaction was studied over the range (0.1-1.0) M. Maximum and constant absorption intensities were achieved at addition of 1.0 mL of 0.2 M HCl, after which the absorbance of the reaction product began to decrease, therefore; 1.0 mL of 0.2 M was chosen as the optimum value, (Figure 3) shows this effect.

Effect of Sulfamic Acid Concentration

The optimum sulfamic acid concentration was estimated by adding 1.0 mL from various concentration (0.1-1.0) % (m/v) of sulfamic acid solution; it was found that 1.0 mL of 0.3 % (m/v) solution give the highest absorbance value. Sulfamic acid was added to remove the excess amount of nitrite; (Figure 4) shows the effect of sulfamic acid concentration.

Effect of Diphenylamine Concentration

Concentration of diphenylamine ranged from (0.25-1.25) % (m/v) of 1.0 mL solutions were examined to obtain highest color intensity of the azo dye as shown in (Figure 5), the investigations showed that 1.0 mL of 0.5 % DPA gave maximum and stable intensity, above this concentration, the absorbance remained unchanged.

Effect of Coupling Reaction Time

The optimum time of coupling reaction time was determined by following the color azo-dye development at room temperature. The reaction mixture was allowed to stand for different intervals, and it was found that 10 minutes period was required for full color development; the color was stable for at least 1h, as indicated in (Table 3).

Effect of Acidity

It was found that the optimum concentration of hydrochloric acid leading to a maximum intensity of the complex was 6 M .Concentrations below and above this value cause a decrease in absorbance; this may be attributed to the de-colorization of colored azo dye. (Figure 6) shows the effect of acidity on color dye.

On the otherhand, the simplex program was employed to find the optimum experimental conditions for determination of (SMZ). The boundary conditions for the three variables delineated above, as recorded in (Table 4) together with the step values .

Four (n+1) arbitrary experimental conditions were chosen, by selecting the values of these parameters within specified boundaries for each, at which they affected the measured absorptions signal of the colored products (1-4 in Table 5) , the absorbencies of these four experiments were fed into the program .

The Simplex program starts to reflect the worst point through the centroid of other points to obtain a new point 5. An experiment was then performed utilizing the variable setting as a reflected point; because this value was better than that at point 3, the latter was rejected and replaced by point 5.

A measured absorption signal was feeding again to the program and the process is repeated successively until optimum conditions were obtained similarly to those obtained by the univariate method. (Figure 7) shows the progress of the simplex, which indicates gradual improvement in the response function. Only 12 experiments were performed, enough to evaluate the proper conditions at maximum response function with the results given in (Table 5) .

Steps 5, 7, 9 and 12 show the same highest response function value obtained. To conclude, the optimum operating conditions for the determination of (SMZ) are 1.0 mL of each 0.20 % (m/v) sodium nitrite, 1.0 % (m/v) DPA and 0.90 M hydrochloric acid.

Calibration Curves and Analytical Data

I. Univariate Method

The effect of concentration on the absorbance behavior at optimum conditions of univariable method was investigated using authentic standard. The results are shown in (Figure 8).

Beer's law was obeyed in the range of 0.5-7.0 $\mu g.mL^{-1}$ of SMZ. The regression equation, correlation coefficient, molar absorptivity, Sandell's sensitivity and detection limit (DOL) and quantification of limit (QOL) are calculated and listed in (Table 6).

II. Simplex Method

Optical characteristics and statistical data for the regression equation of the simplex method are given in Table 7 and Figure 9.

Precision and Accuracy

The accuracy of the both methods were established by analyzing the pure drug at three concentration levels of five replicate analyses and the precision was examined by determining the coefficient of variation (C.V) % on the same solution of drug sample (Table 8). The low values of C.V % (0.139-0.424) and the range of error at the level (-0.500-0.275) indicate the high accuracy and precision of the proposed method.

Interference Study

To assess the analytical potential of the proposed method, the effect of some common excipients; sucrose, vanillin, glucose, lactose, starch, which often accompany drug, were examined by carrying out the determination of 5.0 $\mu g.mL^{-1}$ of SMZ in the presence of above compounds. The results presented in (Table 9), indicate no interferences were found from any of the excipients studied in determination of SMZ.

Application in pharmaceutical preparations by Standard Additions Method (SAM)

Standard additions technique followed to check the validity of the proposed method has given good recoveries of the drug in pharmaceutical preparations suggesting a noninterference from pharmaceutical preparations. Hence, this method can be recommended for adoption in routine analysis of SMZ in quality control laboratories.

(Table 10) shows the result of recovery and coefficient of variation (C.V) for the standard additions method. Figures 10 to 13 show the plot of determination of SMZ in syrup and tablet by standard additions method.

Conclusion

Diazotization reaction of primary amine group followed by coupling with diphenylamine in acidic medium was found to be a simple, sensitive, accurate and economic spectrophotometric method for quantitative determination of (SMZ) in pure form and in pharmaceutical preparation.

The classical univariate and modified simplex method have been used for optimizing the different variable affecting the completion of the reaction. The proposed method offers good linearity and precision.

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Table No. (1): Effect of diazotization reaction time.

Time (min.)	Absorbance
0	0.422
5	0.396
10	0.387
15	0.338
20	0.280

Table No. (2): Effect of different acids on diazotization of (5.0 µg.L^{-1}) SMZ.

Acidic solution (1 M)	Absorbance
H_2SO_4	0.575
HCl	0.663
HNO_3	0.508
CH_3COOH	0.290

Table No. (3): Effect of coupling reaction time.

Time (min.)	Absorbance
0	0.642
5	1.018
10	1.032
15	1.040
20	1.031
60	1.036

Table No. (4): Boundary of simplex for the studied variables.

Variable	Minimum boundary	maximum boundary	Step size
Conc. of sodium nitrite (% m/v)	0.01	0.30	0.01
Conc. of HCl (M)	0.10	1.0	0.10
Conc. of DPA (% m/v)	0.25	1.25	0.25

Table No.(5): Multivariate experiments (simplex) for determination of (5.0 µg.mL^{-1}) SMZ.

Exp. No.	Conc. of sodium nitrite (% m/v)	Conc. of DPA (% m/v)	Conc. Of hydrochloric acid (M)	Abs.
1	0.01	0.25	0.20	0.788
2	0.02	0.40	0.40	0.878
3	0.10	0.60	0.80	0.904
4	0.16	0.75	0.50	0.918
5	0.20	1.00	0.90	0.995
6	0.20	1.00	1.00	0.988
7	0.20	1.00	0.90	0.995
8	0.18	0.90	0.60	0.978
9	0.20	1.00	0.90	0.995
10	0.19	1.00	0.90	0.975
11	0.20	1.00	0.80	0.970
12	0.20	1.00	0.90	0.995

Table No. (6): Optical characteristics and statistical data for determination of SMZ by univariate method.

Parameter	Value
λ_{max} (nm)	530.0
Color	Blue
Linearity range (µg.mL^{-1})	0.5-7.0
Regression equation	Y= 0.2043[SMZ. µg.mL^{-1}]-0.001
Calibration sensitivity (L. µg^{-1}.cm^{-1})	0.2043
Correlation coefficient (r)%	99.98
Correlation of linearity (r^2)%	99.97
Molar absorptivity (L.mol^{-1}.cm^{-1})	$\varepsilon = 5.1745 \times 10^4$
Sandell's sensitivity (µg.cm^{-2})	4.8947
Detection limit (µg.mL^{-1})	0.034
Quantification limit (µg.mL^{-1})	0.104

Table No. (7) : Optical characteristics and statistical data for determination of SMZ by simplex method.

Parameter	Value
λ_{max} (nm)	530.0
Color	Blue
Linearity range ($\mu g.mL^{-1}$)	0.5-12.0
Regression equation	Y=0.1959[SMZ. $\mu g.mL^{-1}$]-0.0083
Calibration sensitivity (L. $\mu g^{-1}.cm^{-1}$)	0.1959
Correlation coefficient (r)%	99.99
Correlation of linearity (r^2)%	99.98
Molar absorptivity (L.mol $^{-1}$.cm $^{-1}$)	$\varepsilon = 4.9617 \times 10^4$
Sandell's sensitivity ($\mu g.cm^{-2}$)	5.1047
Detection limit ($\mu g.mL^{-1}$)	0.036
Quantification limit ($\mu g.mL^{-1}$)	0.108

Table No. (8): Evaluation of accuracy and precision for the determination of SMZ by proposed method.

	Conc. of SMZ ($\mu g.mL^{-1}$)		Relative Error %	C.V %
	Taken	Found*		
For univariate	2.000	1.989	-0.550	0.190
	4.000	4.011	0.275	0.424
	6.000	6.014	0.233	0.369
For simplex	2.000	1.990	-0.500	0.139
	4.000	4.010	0.250	0.233
	6.000	6.013	0.216	0.224

*Average of five determinations.

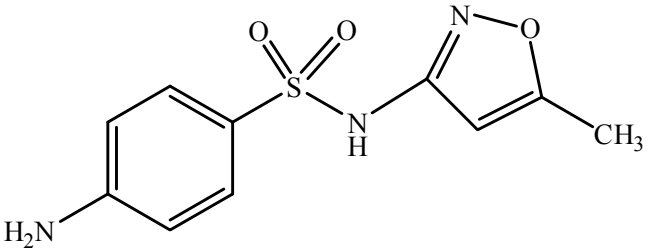
Table No. (9): Percent recovery for (5.0 $\mu g.mL^{-1}$) of sulfamethoxazole in the presence of (1000 $\mu g.mL^{-1}$) of excipients.

Excipients	Concentration of Excipient ($\mu g.mL^{-1}$)	Sulfamethoxazole Conc. Taken 5.0 ($\mu g.mL^{-}$	
		Conc. Found ($\mu g.mL^{-1}$)	Recovery %
Sucrose	1000	5.02	100.40
Vanillin		4.94	98.80
Glucose		4.96	99.20
Lactose		4.92	98.40
Starch		5.06	101.20

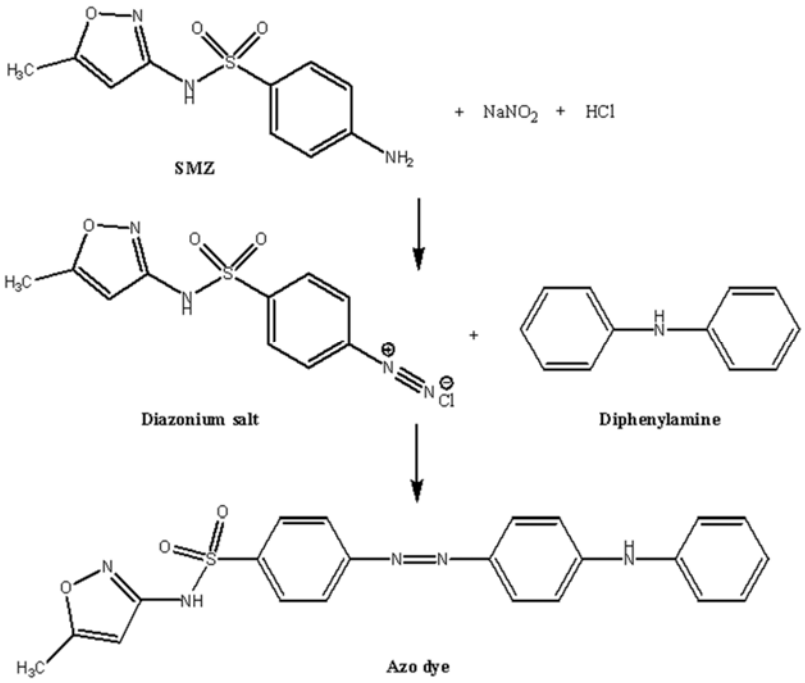
Table No. (10): Application of the proposed method to the SMZ concentration measurements pharmaceutical preparations by (SAM).

Sample	Conc. taken ($\mu g.mL^{-1}$)	Conc.* found ($\mu g.mL^{-1}$)	Recovery %	C.V* %
Bactrim tablet (Roche-France)	100.00	101.33	101.33	0.442
Bactrim syrup (Roche-France)	100.00	100.13	100.13	0.443
Methoprim tablet (NDI-Iraq)	100.00	96.54	96.54	0.867
Cotrim syrup (Asia-Syria)	100.00	101.00	101.00	0.469

*Average of three determinations.



Scheme No. (1): The chemical structure of sulfamethoxazole.



Scheme No. (2): The reaction mechanism for diazotization and reaction between SMZ and DPA.

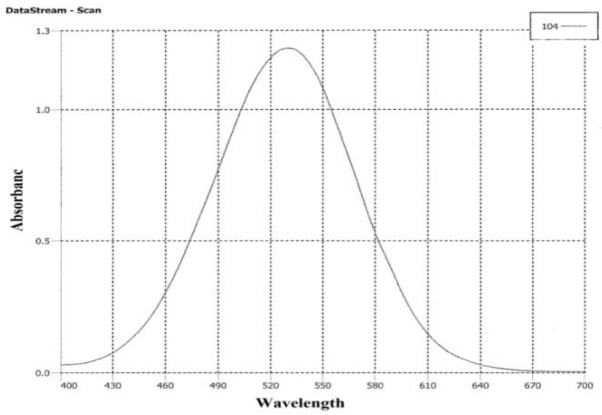


Figure No. (1): Absorption spectrum of (5.0 µg.mL^{-1}) SMZ-DPA against reagent blank under the optimum conditions.

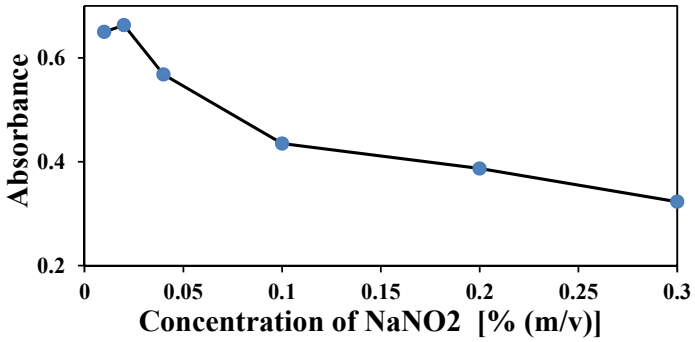


Figure No. (2): Effect of sodium nitrite on the color development of dye on the estimation of (5 µg.mL^{-1}) SMZ.

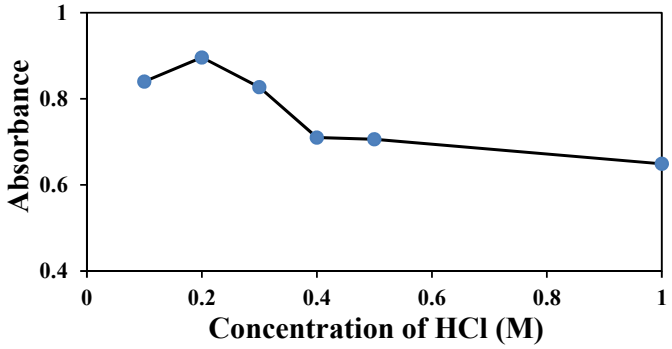


Figure No. (3): Effect of acidity on diazotization on the estimation of (5.0 µg.mL^{-1}) SMZ.

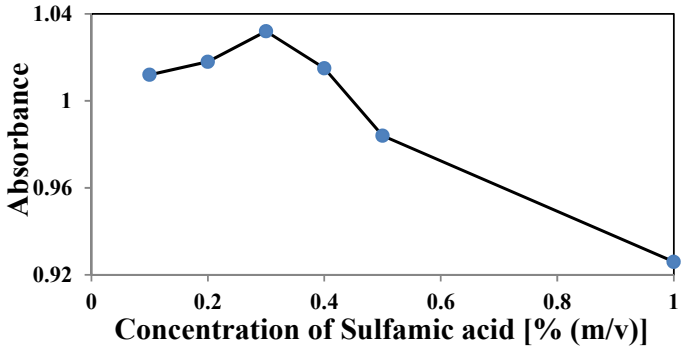


Figure No.(4): Effect of sulfamic acid concentration on the estimation of (5.0 µg.mL^{-1}) SMZ.

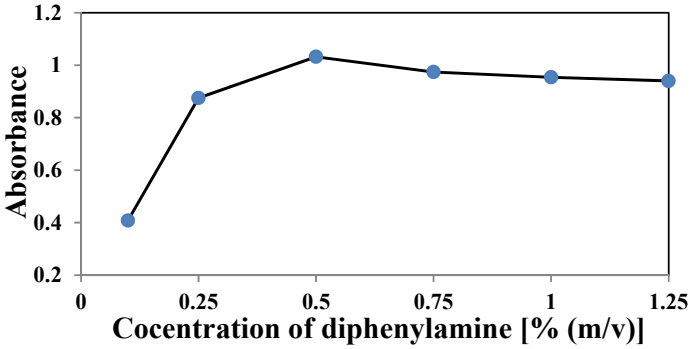


Figure No. (5): Effect of DPA concentration on the development of dye on the estimation of (5.0 µg.mL^{-1}) SMZ.

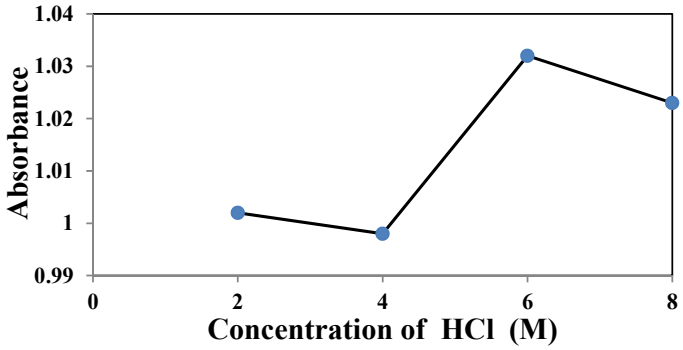


Figure No. (6): Effect of acidity on color dye on the estimation of (5.0 µg.mL^{-1}) SMZ.

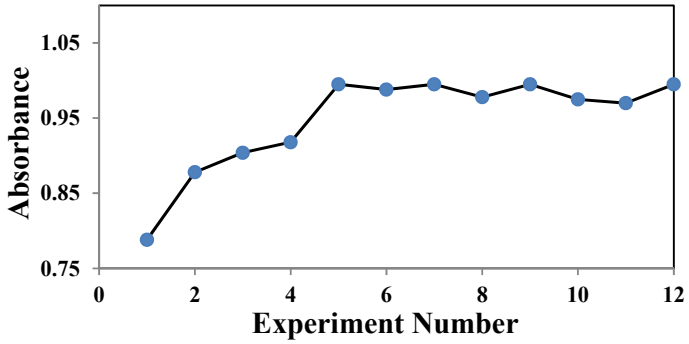


Figure No. (7): Response function progress for simplex.

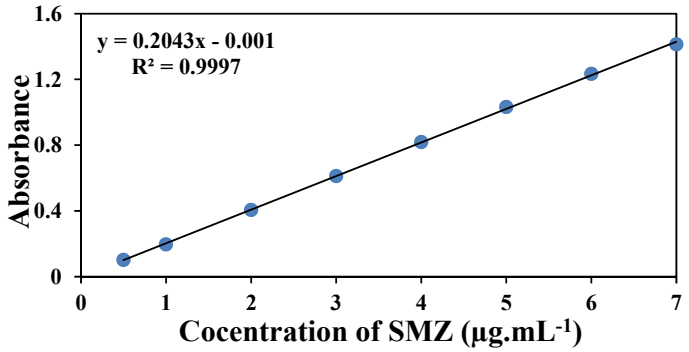


Figure No. (8): Calibration curve for determination of SMZ by univariate optimal condition.

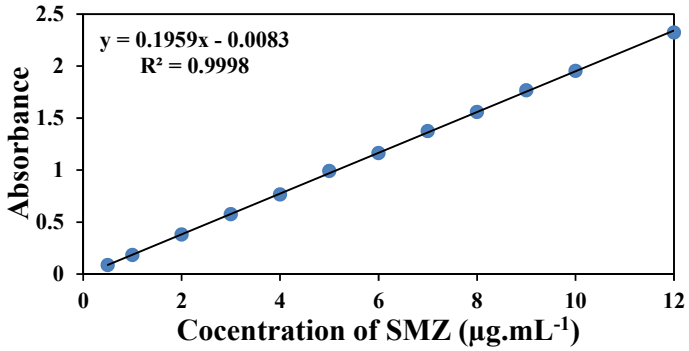


Figure No.(9): Calibration curve for determination of SMZ by simplex optimal condition.

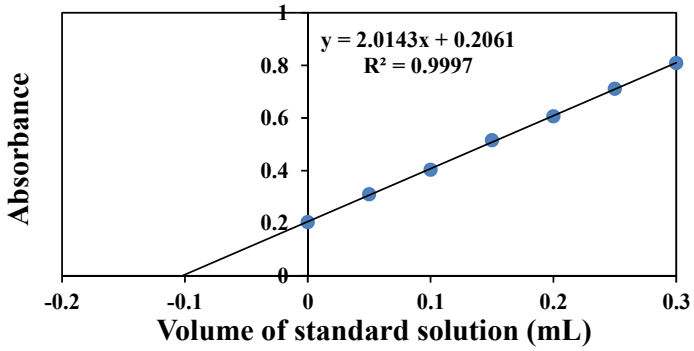


Figure No. (10): Determination of SMZ in (Bactrim) tablet by standard additions method level one, (0.1 mL from 100 µg.mL^{-1} Bactrim tablet).

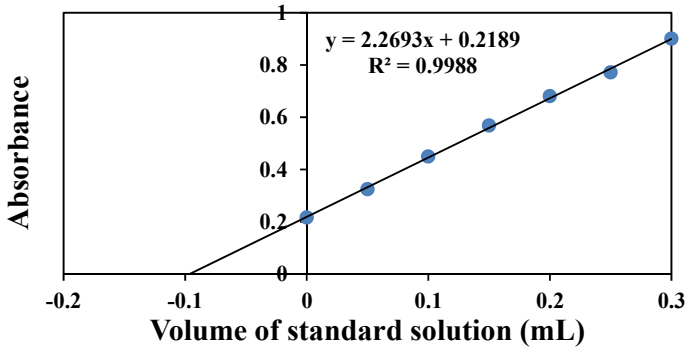


Figure No. (11): Determination of SMZ in (Methoprim) tablet by standard additions method level one, (0.1 mL from 100 µg.mL^{-1} Methoprim tablet).

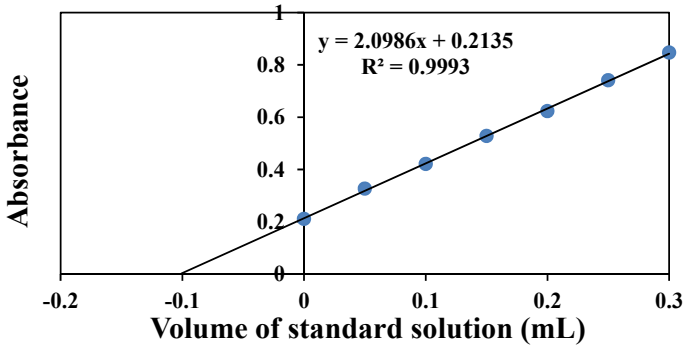


Figure No. (12): Determination of SMZ in (Bactrim) syrup by standard additions method level one, (0.1 mL from 100 µg.mL^{-1} Bactrim syrup).

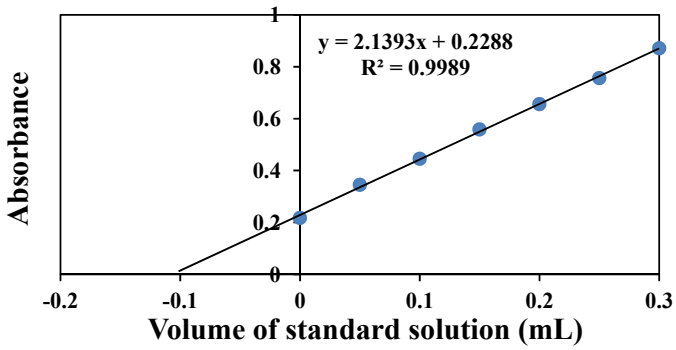


Figure No. (13): Determination of SMZ in (Cotrim) syrup by standard additions method level one, (0.1 mL from 100 µg.mL^{-1} Cotrim syrup).

التقدير الطيفي للسلفاميثوكسازول المتضمن الازوتة البسيطة والازدواج مع داي فينيل امين

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استلم البحث في: 7ايار2014,قبل في 22حزيران2014

الخلاصة

طورت طريقة طيفية بسيطة وحساسة ودقيقة وغير مكلفة للتقدير الكمي للسلفاميثوكسازول بشكله النقي وفي المستحضرات الصيدلانية. تعتمد الطريقة على ازوتة مجموعة الامين الاولي في السلفاميثوكسازول بمعاملته مع نتريت الصوديوم وحامض الهيدروكلوريك ثم اجراء تفاعل الازدواج مع داي فينيل امين في وسط حامضي للحصول على كروموجين مستقر ذي لون أزرق يظهر أعظم امتصاص عند 530 نانومتر. وقد تم دراسة العوامل التي تؤثر في اتمام التفاعل بعناية للحصول على الظروف المثلى وذلك بأتباع نمط المتغير الواحد وبالاعتماد على طريقة السمبلكس المحورة. عند الظروف المثلى، وجد ان قانون بير ينطبق على مدى من التراكيز يتراوح بين ($0.5 - 12.0\ \mu g.mL^{-1}$) مع قيمة امتصاصية مولارية مساوية ل ($4.9617\times10^{4}\ L.mol^{-1}.cm^{-1}$) وكان حد الكشف يساوي ($0.108\ \mu g.mL^{-1}$) ودلالة ساندل تساوي ($4.8947\ \mu g.cm^{-2}$). لقد أمكن تطبيق الطريقة المقترحة بنجاح لتقدير السلفاميثوكسازول في المستحضرات الصيدلانية.

الكلمات المفتاحية: التقدير الطيفي، الاشكال الصيدلانية، سلفاميثوكسازول، تفاعل الازوتة، تفاعل الازدواج، داي فينيل أمين.