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SIMULTANEOUS ESTIMATION AND VALIDATION OF KETOCONAZOLE AND SALICYLIC ACID IN BULK AND EMULGEL FORMULATION BY SPECTROPHOTOMETRIC METHODS

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ABSTRACT

Two simple and economic UV Visible spectrophotometric methods have been validated for the determination of Ketoconazole and Salicylic acid in bulk and emulgel formulation. The developed emulgel containing Ketoconazole and Salicylic acid was formulated for the management of Psoriasis. Quantitation was carried out by two spectrophotometric methods such as Simultaneous equation method and Absorbance ratio method. The wavelength selected for Simultaneous equation method were 225 nm (λ_{max} of ketoconazole) and 296 nm (λmax of salicylic acid). In Absorbance ratio method, two wavelength 296 nm, λ_{max} of Salicylic acid as λ_2 and 239 nm, iso-absorptive point as λ_1 was selected. Both the method follows beer's linearity in the range of 3-21 µg/ml for Ketoconazole and 3-21 µg/ml for Salicylic acid with correlation coefficient value (r^2) for both the drug being more than 0.995. According to ICH guidelines the parameters Linearity, Precision, Accuracy, Limit of detection, Limit of quantification, Robustness, Ruggedness was performed. Result of analysis were validated statistically. All the validation parameters were found to be within limit of acceptance.

KEYWORDS: Emulgel, Ketoconazole, Salicylic acid, Psoriasis, Simultaneous Equation Method, Q-Absorption Ratio Method, Validation.

INTRODUCTION

Ketoconazole

Ketoconazole is white coloured powder. It is identified as (cis-1-acetyl-4[4[[2-(2,4-dichlorophenyl)-2-(1H-imidazol-1-ylmethyl)-1,3-dioxalan-4-yl]methoxy]phenyl]piperazine) in (figure 1).[1] Pharmaceutical formulation of Ketoconazole includes creams, tablets and shampoo are used for treatment and prevention of systemic skin infection.[2] Ketoconazole is classified in the biopharmaceutics classification system (BSC) as a class 2 drug. Ketoconazole has a good permeability. It has very low aqueous solubility because of its hydrophobic structure.[3] There are different analytical method for determination of Ketoconazole in single and combine pharmaceutical dosage form.[4,5] Ketoconazole was approved in 1981 by united states food and drug administration (USFDA) for skin infections as an oral anti-fungal agent, later it was withdrawn due to serious adverse events such as drug-drug interaction, hepatotoxicity etc. Further, topical Ketoconazole preparations took place for treating against dermatological infections. This result decrease in drug related adverse effects.[6]

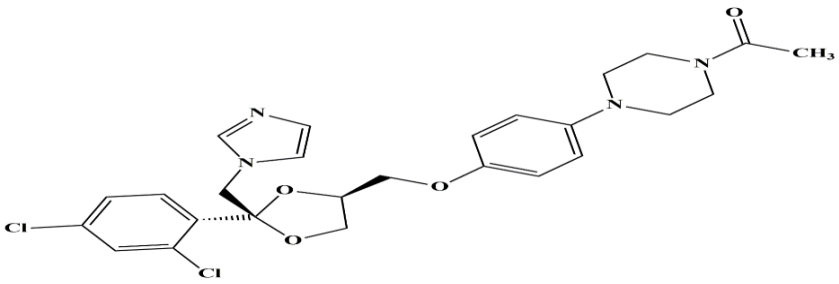

Figure 1: Structure of ketoconazole.

Category: Anti-fungal and used in treating skin plaques

Melting Point: 148℃

Molecular Formula: $C_{26}H_{28}Cl_2N_4O_4$

Molecular Weight: 531.431g/mol

Salicylic acid

Salicylic Acid is chemically, 2-hydroxybenzoic acid as shown in (figure 2). It has activity as an anti-inflammatory agent.[7,8] It exerts anti-inflammatory effect by suppressing cyclooxygenase activity. It is use in treatment of various skin disorders like acne, psoriasis, seborrheic dermatitis, calluses, keratosis pilaris and warts. Salicylic acid is mostly used in

topical formulations.[9] Salicylic Acid is white, crystalline powder needle. Salicylic Acid prepared from natural salicylate may have slightly yellow or pink color. It is stable in air.[10]

Salicylic Acid has the ability to exfoliate stratum corneum which makes it a good peeling agent for acne patients. It is effective and safe for dermatological preparation. Salicylic acid absorbs readily when applied to the skin. It is toxic in higher concentration.[11]

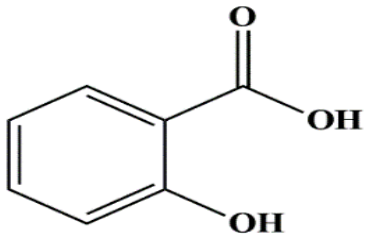

Figure 2: Structure of salicylic acid.

Category: Anti-inflammatory
Melting Point: 157-159℃
Molecular Formula: $C_7H_6O_3$
Molecular Weight: 138.12 g/mol

MATERIAL AND METHODS

Instruments

The experimental work was executed using instruments such as a double beam UV-Vis Spectrophotometer (Model, SHIMADZU UV-1900) with a spectral band width of 1nm and automatic wavelength correction, as well as a pair of 10mm quartz cells, Digital Weighing Balance (Mettler Toledo), Sonicator (Inco) and pH Meter

Materials

Gift sample of Ketoconazole was procured from Aarti Drugs Pharmaceutical Pvt. Ltd. Boisar, and Salicylic Acid was procured from Dales Remedies Pvt. Ltd. Palghar. All chemicals were of analytical grade.

Preparation of stock solution

Standard stock solution of both Ketoconazole and Salicylic acid were prepared separately, dissolving 100 mg of each drug in 100 ml volumetric flask and made up with Ethanol and Buffer pH-5 in ratio 1:1 to obtain stock solution of 1000 µg/ml.

Determination of λ_{max}

The standard stock solution of Ketoconazole (15 µg/ml) and Salicylic acid (15 µg/ml) were scanned separately in the wavelength ranges from 200-400 nm and the λ_{max} was found to be 225 nm of Ketoconazole and 296 nm of Salicylic Acid in ethanol and buffer pH-5 in ratio 1:1 solvent as expressed in (figure 3A and 3B).

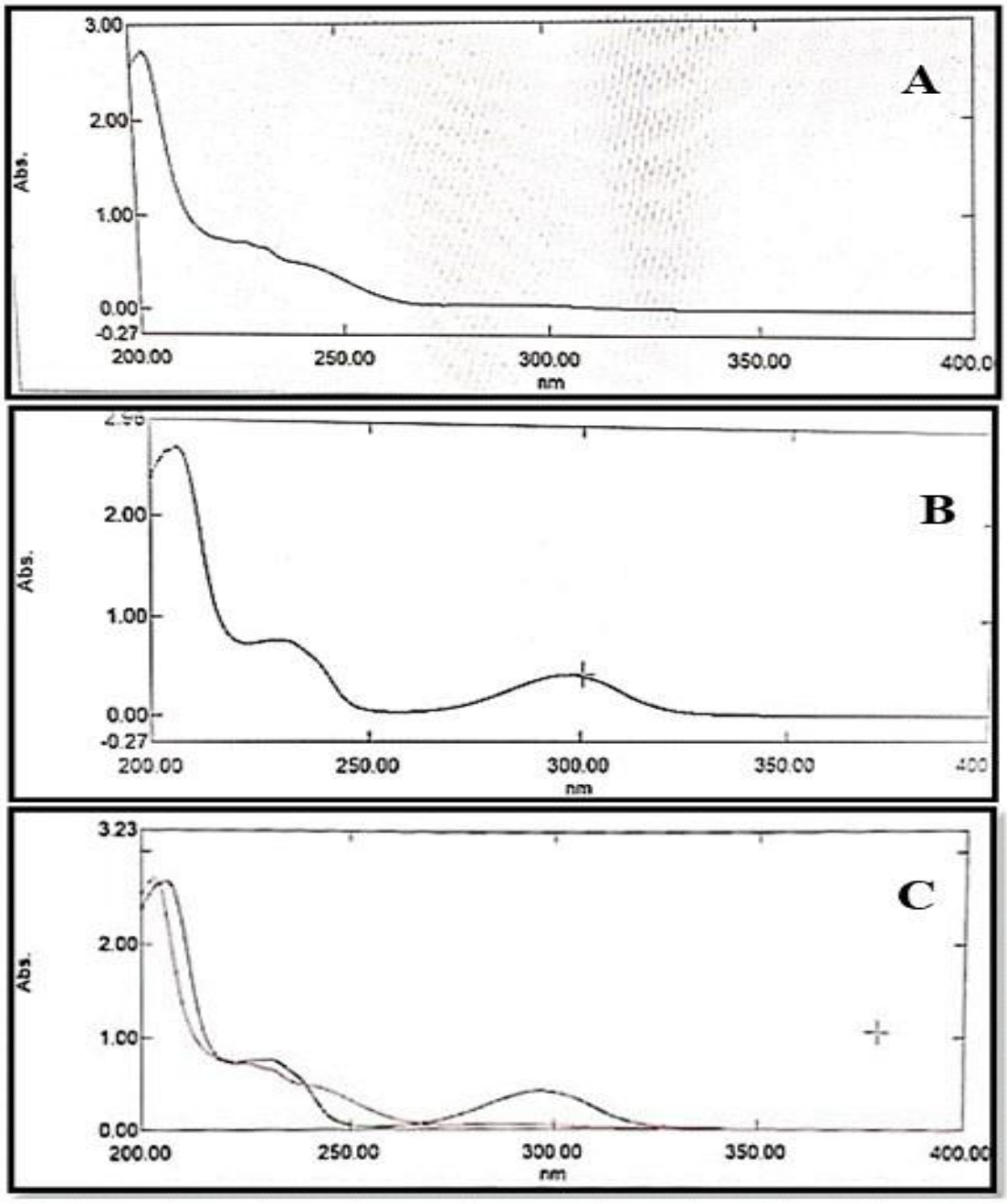

Figure 3: (A) UV Spectra of Ketoconazole at 225 nm (15 µg/ml) (B) UV spectra of Salicylic acid at 296 nm (15 µg/ml (C) Overlay spectra of Ketoconazole and Salicylic acid.

Methods

Both drugs overlay at the wavelength 239 nm and according to overlain spectra of Ketoconazole and Salicylic Acid two methods have been carried out for estimation of both the drugs i.e., Simultaneous equation method and Absorbance ratio method as (figure 3C).

Method I: Simultaneous equation method[12,13]

15 µg/ml solution of Ketoconazole and Salicylic Acid was prepared separately in phosphate buffer pH-5 and ethanol in ratio 1:1 and the solution were scanned against blank in the range of 200-400 UV range to determine λ_{max} values.

Peaks were observed at 225 nm for Ketoconazole and 296 nm for Salicylic Acid. These wavelengths were selected as λmax of each drug. Standard solution of both the drugs in concentration range 3-21 µg/ml for both the drugs were prepared in buffer solution of pH-5 and ethanol in ratio 1:1 and absorbance of these solution was measured at 225 nm and 296 nm. Calibration curve were plotted to verify the beer's law.

The concentration of both the drugs in mixture was calculated by using following equations.

$$C_X = (A2 \times ay1 - A1 \times ay2) / (ax2 \times ay1 - ax1 \times ay2)$$

$$C_Y = (A1 \times ax2 - A2 \times ax1) / (ax2 \times ay1 - ax1 \times ay2)$$

Where,

C_X and C_Y =Concentration of Ketoconazole and Salicylic acid in mixture

A1 and A2 =Absorbance of mixture at 225 nm and 296 nm

ax1 and ax2 = Absorptivities of Ketoconazole at 225 nm and 296 nm

ay1 and ay2 = Absorptivities of Salicylic acid at 225 nm and 296 nm

Method II: Q-Absorbance ratio method[14-18]

Absorbance ratio method depends on property, which obeys Beer's law at all wavelength, the ratio of absorbance at any two wavelengths is constant value independent of path length or concentration. This ratio is referred to as "Hufner's Quotient" or Q-value.

In absorbance ratio method, absorbances are measured at two wavelengths, one being a wavelength of equal absorptivity of the two components (λ_1) i.e., an iso- absorptive point. From the overlay spectra of both drugs (figure 5), two iso-absorptive point were detected as 239 nm and 267 nm. But at 267 nm very less absorbance was recorded, so 239 nm was selected as λ_1 and other being λ_{max} of one of the components λ_2.

15µg/ml solution of Ketoconazole and Salicylic Acid was prepared separately in phosphate buffer pH-5 and ethanol in ratio 1:1 and the solution were scanned against blank in the range of 200-600 UV range to determine λ_{max} values.

Standard solution of both the drugs in concentration range 3-21 µg/ml for both the drugs were prepared in buffer solution of pH-5 and ethanol in ratio 1:1 and absorbance of these solution was measured at 239 nm (iso-absorptive point) and 296 nm selected wavelength (λ_{max} of Salicylic Acid).

Concentration of individual components, C_X and C_Y Can be calculated by using the following equation.

$$C_X = Q_M - Q_y / Q_x - Q_y \times A_1 / ax_1$$

$$C_Y = Q_M - Q_y / Q_X - Q_y \times A_1 / ay_1$$

Where,

A_1= Absorbance of mixture at iso-absorptive point (239 nm)

Q_M= Absorbance of mixture at selected wavelength (296 nm) divided by absorbance of mixture at iso-absorptive point.

Q_X and Q_Y =Deviation of respective drugs absorbance at selected wavelength 296 nm and iso-absorptive point.

ax_1 and ay_1= Absorbance of Ketoconazole and Salicylic Acid at iso-absorptive point divided by concentration of respective drug.

Validation of Uv-visible spectrophotometric methods

The developed method was validated according to the International Conference on Harmonization (ICH) Guidelines.[19-22]

Linearity and Range[13]

Seven aliquots were taken from standard stock solution of both the drugs Ketoconazole and Salicylic acid and transferred to 10 ml volumetric flask to get a concentration of 3, 6, 9, 12, 15, 18, 21 µg/ml of Ketoconazole and 3, 6, 9, 12, 15, 18, 21 µg/ml of Salicylic acid in triplicate. The volume was made up with phosphate buffer pH-5 and ethanol in ratio 1:1 and absorbance were measured at selected wavelengths.

For Simultaneous equation method the absorbance of all solutions was measured at 225 nm of Ketoconazole and 296 nm of Salicylic acid. The calibration curve of Absorbance vs.

Concentration was plotted with correlation coefficient and regression line equation for both the drug was determined.

For Q-Absorbance ratio-method the wavelength selected was 239 nm (Iso-absorptive) and 296 nm selected wavelength (λ_{max} of Salicylic acid). The absorbance of all the solutions at these two wavelengths of both Ketoconazole and Salicylic acid was measured and calibration curve and linear regression equation was determined.

Accuracy (Recovery)

Accuracy of the methods was done by recovery study from optimized formulation at three level of standard addition (80%, 100%, 120%). Fixed concentration of the developed emulgel formulation, with varying concentration of pure drug solution were added and percent recoveries was calculated. For Simultaneous equation method absorbance was measured at 225 nm for Ketoconazole and at 296 nm for Salicylic acid at 80%, 100%, 120% of the standard preparation in ratio of the formulation was prepared and checked for accuracy. For Absorbance ratio method absorbance were measured at 296 nm and 239 nm for both the drug Ketoconazole and Salicylic acid followed as same as procedure for accuracy.

Precision

Intra-day study was performed by analyzing response of three different concentration (9, 12, 15 µg/ml) three times on same day at an interval of one hour. While Inter-day study was performed by analyzing response of same concentrations three times on three different days over a period of one week.

Results was expressed in terms of % RSD for both the drugs Ketoconazole and Salicylic acid in triplicate.

Limit of Detection (LOD) and Limit of quantification (LOQ) [23]

LOD and LOQ was calculated from linearity studies. From regression equation slope was determined and standard deviation was computed for both the drug Ketoconazole and Salicylic acid. From these values, LOD and LOQ was determined by equation.

LOD= 3.3*σ/S

LOQ= 10*σ/S

σ =Standard deviation

S= slope of calibration curve

Robustness

Robustness of the proposed method was determined by changing λ_{max} by ± 2nm for both the drugs and results were obtained at different wavelength and %RSD was reported.

For Simultaneous Equation variation in λ_{max} of Ketoconazole at 225 nm with ± 2nm was carried out for 15 µg/ml in triplicate similarly for λ_{max} of Salicylic acid at 296 nm with ± 2nm was carried out. For Absorbance ratio method variation in iso-absorptive point 239 nm by ± 2nm and λ_{max} of Salicylic acid 296 nm (selected wavelength) by ± 2 nm was carried out for 15 µg/ml concentration of Ketoconazole and Salicylic acid in triplicate.

Ruggedness

Ruggedness was determined for 15 µg/ml concentration of both the drug Ketoconazole and Salicylic acid by two analysts using same operational and environmental conditions. Result was indicated as %RSD. For Method I absorbance was carried out at 296 nm of Salicylic acid and 225 nm of Ketoconazole by analyst 1 and analyst 2. For Method II absorbance was carried out at 239 nm (iso-absorptive point) and (selected wavelength) 296 nm of Salicylic acid by analyst 1 and analyst 2.

RESULTS

Linearity and Range

The linearity of Ketoconazole and Salicylic acid was found to be in range 3-21µg/ml in Method I and Method II. For Method I, the correlation coefficient for Keto at 225 nm and SA at 296 nm is 0.9989 and 0.9991 respectively which expressed in (figure 4). Linear regression equation was found to be Y= 0.0439x - 0.0027 and Y= 0.026x + 0.0059. For Method II, the correlation coefficient 0.9991 and 0.999 for Ketoconazole and 0.9991 and 0.9991 for Salicylic Acid at 239 nm and 296 nm expressed in (figure 5). Linear regression equation for Ketoconazole was found to be Y= 0.0298x + 0.0008 and Y = 0.0037x + 0.001 and for Salicylic Acid was found to be Y = 0.032x + 0.0079 and Y = 0.026x + 0.0059. Linearity data are reported in (Table 1 (Method I) and Table 2 (Method II)). Optical and regression parameters are summarized in (Table 3)

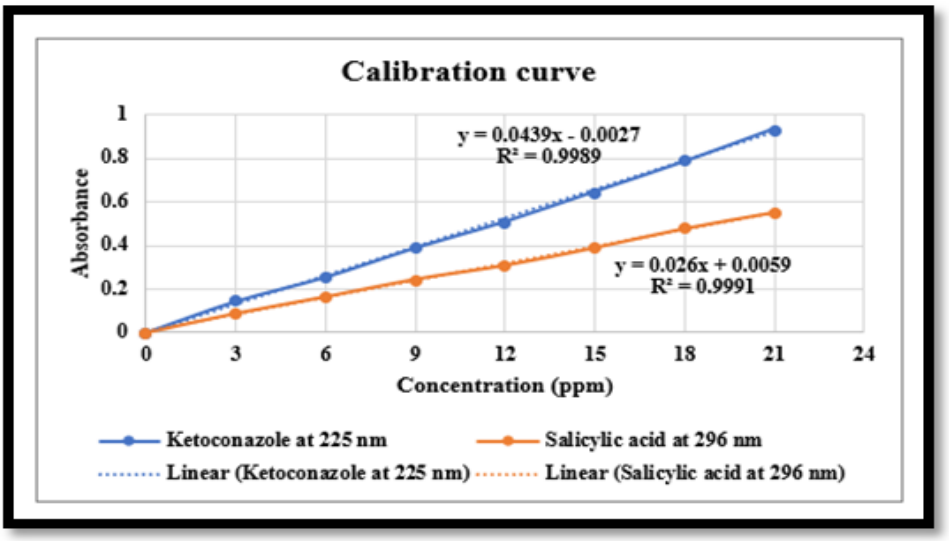

Figure 4: Calibration curve of ketoconazole at 225 nm and Salicylic acid at 296 nm.

Table 1: Calibration data of Ketoconazole and Salicylic acid for method I (in triplicate).

Sr. no.	Conc (µg/ml)	Ketoconazole at 225 nm		Salicylic acid at 296 nm	
		Absorbance*mean ± SD	%RSD	Absorbance*mean ± SD	%RSD
1.	0	0	0	0	0
2.	3	0.1433 ± 0.0025	1.75	0.089 ± 0.001	1.12
3.	6	0.2547 ± 0.0031	1.19	0.16533 ± 0.0025	1.52
4.	9	0.389 ± 0.0026	0.68	0.246 ± 0.002	0.81
5.	12	0.5097 ± 0.0021	0.40	0.30967 ± 0.0015	0.49
6.	15	0.6477 ± 0.0015	0.23	0.39033 ± 0.004	1.03
7.	18	0.7857 ± 0.0035	0.44	0.47833 ± 0.0031	0.63
8.	21	0.936 ± 0.0036	0.38	0.55233 ± 0.0025	0.45

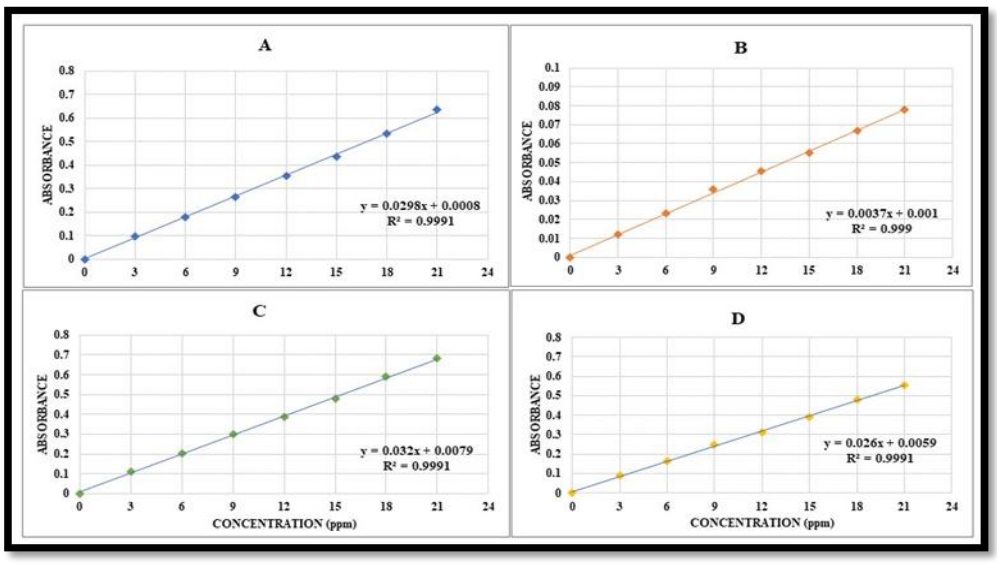

Figure 5: Calibration curve of (A) Ketoconazole at 239 nm (B) Ketoconazole at 296 nm (C) Salicylic acid at 239 nm (D) Salicylic acid at 296 nm.

Table 2: Calibration data of Salicylic acid and Ketoconazole for Method II (in triplicate).

Sr. no.	Salicylic acid							Ketoconazole					
	Concentration (µg/ml)	Absorbance at 239 nm			Absorbance at 296 nm			Absorbance at 239 nm			Absorbance at 296 nm		
		Mean (n=3)	SD	% RSD	Mean (n=3)	SD	% RSD	Mean (n=3)	SD	% RSD	Mean (n=3)	SD	% RSD
1.	0	0	0	0	0	0	0	0	0	0	0	0	0
2.	3	0.112	0.0015	1.356	0.089	0.001	1.12	0.098	0.001	1.55	0.012	0.001	8.33
3.	6	0.204	0.002	0.98	0.1653	0.0025	1.52	0.179	0.003	1.67	0.023	0.006	2.4
4.	9	0.3	0.002	0.667	0.246	0.002	0.81	0.266	0.003	1.35	0.035	0.006	1.66
5.	12	0.385	0.002	0.653	0.309	0.0015	0.49	0.354	0.002	0.74	0.045	0.006	1.26
6.	15	0.477	0.002	0.419	0.390	0.004	1.03	0.436	0.002	0.57	0.055	0.001	1.81
7.	18	0.589	0.002	0.353	0.478	0.0031	0.63	0.535	0.002	0.38	0.067	0.001	1.49
8.	21	0.682	0.002	0.369	0.552	0.0025	0.45	0.636	0.003	0.47	0.078	0.001	1.28

Table 3: Optical and regression parameter of the calibration curve obtained by UV spectroscopy methods (Average of 3 determinations).

Parameters	Method 1		Method 2			
	Ketoconazole	Salicylic acid	Ketoconazole	Salicylic acid	Ketoconazole	Salicylic acid
Wavelength λmax (nm)	225	296	Isosbestic point at 239 nm		Selected Wavelength at 296 nm	
Beer's law limit (µg/ml)	3-21	3-21	3-21	3-21	3-21	3-21
Correlation coefficient r^2	0.9989	0.9991	0.9991	0.9991	0.9989	0.9991
Slope	0.0439	0.026	0.0298	0.032	0.0037	0.026
Intercept	0.0027	0.0059	0.0008	0.0079	0.0011	0.0059
Intra-day (%RSD)	0.967	0.717	0.473	1.486	1.20	0.717
Inter-day (%RSD)	1.710	0.777	1.683	1.547	1.660	0.777
LOD (µg/ml)	0.4747	0.7047	0.6783	0.5033	1.8015	0.7047
LOQ (µg/ml)	1.4384	2.1355	2.0556	1.5253	5.4591	2.1355

Accuracy (Recovery study)

For Method I, % recoveries were found to be 99.8-102.7% for Ketoconazole and 82.9-92.81% for Salicylic acid. For Method II, % recoveries were found to be 96.03-98.8% (296 nm) and 87.7-97.6% (239 nm) for Ketoconazole and 102.6-108% (296 nm) and 97-100% (239 nm) for Salicylic acid. The recovery studies are reported in (Table 4**).**

Table 4: Result of % Recovery and % RSD of Ketoconazole and Salicylic acid for Method I and Method II (*Average of 3 determinations).

Drugs	Recovery level	Initial conc. (Test-formulation) µg/ml	Conc. of Std. Drug added µg/ml	Method I		Method II			
				% Recovery	% RSD	% Recovery		% RSD	
				λ_{max}		296 nm	239 nm	296 nm	239 nm
Keto	80	6	4.8	99.8	1.73	97	87.7	5.26	1.99
	100	6	6	102.7	1.49	96.03	97.6	2.47	1.13
	120	6	7.2	100	0.65	98.8	92.3	3.44	1.52
SA	80	6	4.8	83.1	1.39	102.6	97	1.4	1.75
	100	6	6	82.9	1.53	106	98	1.16	0.77
	120	6	7.2	92.81	1.46	108	100	0.98	1.27

Precision

For Method I, % RSD of intra-day and inter-day precision was found to be less than 2 for both the drugs. For Method II, % RSD for Intra-day and inter-day precision of both Ketoconazole and Salicylic acid was found to be less than 2 at 239 nm and 296 nm. Results are expressed in (Table 5).

Table 5: Result of Intra-day and Intra-day precision of Ketoconazole and Salicylic acid for Method I and Method II.

Sr. No.	Conc. (µg/ml)		λ_{max} (nm)	Intra-Day Absorbance*mean ± S.D.		% RSD		Inter-Day Absorbance*mean ± S.D.		% RSD	
	Keto	SA		Keto	SA	Keto	SA	Keto	SA	Keto	SA
Method I											
1.	9	9	Keto-225 nm & SA-296 nm	0.3837 ± 0.0047	0.2481 ± 0.0018	1.2	0.76	0.3954 ± 0.0072	0.2444 ± 0.0016	1.83	0.68
2.	12	12		0.5078 ± 0.0016	0.3122 ± 0.0030	0.3	0.99	0.5076 ± 0.0101	0.3097 ± 0.0003	1.99	0.10
3.	15	15		0.6368 ± 0.0095	0.3921 ± 0.0015	1.4	0.40	0.6554 ± 0.0086	0.3964 ± 0.0061	1.31	1.55
Method II											
1.	9	9	239	0.2613 ± 0.004	0.3016 ± 0.0015	0.50	1.59	0.2603 ± 0.0051	0.2952 ± 0.0042	1.97	1.42

			296	0.0337 ± 0.0003	0.2481 ± 0.0019	0.76	1.11	0.0343 ± 0.0006	0.2444 ± 0.0017	1.82	0.68
2.	12	12	239	0.3480 ± 0.005	0.3863 ± 0.0018	0.45	1.59	0.3489 ± 0.004	0.3774 ± 0.0072	1.35	1.91
			296	0.0452 ± 0.0003	0.3122 ± 0.0031	0.99	0.87	0.0445 ± 0.0005	0.3096 ± 0.0003	1.20	0.10
3.	15	15	239	0.4296 ± 0.005	0.4795 ± 0.0023	0.47	1.28	0.4313 ± 0.007	0.475 ± 0.0062	1.73	1.31
			296	0.0543 ± 0.054	0.3921 ± 0.0016	0.40	1.62	0.0509 ± 0.001	0.3964 ± 0.0062	1.96	1.55

LOD and LOQ

For Method I, Limit of detection was found to be 0.4747 and 0.7047 for Ketoconazole and Salicylic acid. Limit of Quantification was found to be 1.4384 and 2.1355 for Ketoconazole and Salicylic acid. For Method II, Limit of detection was found to be 0.6783 at 239 nm, 1.8015 at 296 nm for Ketoconazole and 0.5033 at 239 nm, 0.7047 at 296 nm for Salicylic acid. Limit of quantitation was found to be 2.0556 at 239 nm, 5.4591 at 296 nm for Ketoconazole and 1.5253 at 239 nm, 2.1355 at 296 nm for Salicylic acid.

Robustness

In robustness results are obtained at different wavelengths. For Method I and Method II, % RSD for Ketoconazole and salicylic acid at different wavelength was found to be less than 2. Results are expressed in (Table 6 and Table 7)

Table 6: Result of robustness of Ketoconazole and Salicylic acid for method I.

Sr. no.	Drugs	Keto			SA		
	Conc (µg/ml)	223	225	227	294	296	298
1.	15	0.665	0.666	0.665	0.377	0.385	0.384
2.	15	0.662	0.663	0.661	0.38	0.389	0.386
3.	15	0.661	0.662	0.661	0.381	0.391	0.388
4.	Mean	0.6626	0.6636	0.6623	0.3793	0.3883	0.386
5.	SD	0.0021	0.0021	0.0023	0.0020	0.0030	0.002
6.	%RSD	0.3141	0.3137	0.3487	0.5487	0.7867	0.5181

Table 7: Result of robustness of Ketoconazole and Salicylic acid for Method II.

Sr. No.	Drugs	Keto (at selected wavelength)			Keto (at isosbestic wavelength)			SA (at selected wavelength)			SA (at isosbestic wavelength)		
	Conc (µg/ml)	294	296	298	237	239	241	294	296	298	237	239	241
1.	15	0.056	0.055	0.377	0.385	0.377	0.385	0.377	0.385	0.384	0.55	0.469	0.351
2.	15	0.056	0.054	0.38	0.389	0.38	0.389	0.38	0.389	0.386	0.555	0.472	0.353

3.	15	0.057	0.056	0.381	0.391	0.381	0.391	0.381	0.391	0.388	0.556	0.475	0.356
4.	Mean	0.056	0.055	0.3793	0.3883	0.3793	0.3883	0.3793	0.3883	0.386	0.5536	0.472	0.3533
5.	SD	0.000	0.001	0.0021	0.0030	0.0021	0.0030	0.0021	0.0030	0.002	0.00321	0.003	0.0025
6.	% RSD	1.024	1.818	0.5488	0.7867	0.5488	0.7867	0.5488	0.7867	0.5181	0.5806	0.6356	0.7122

Ruggedness

In both the Methods results are in the satisfactory range for Ketoconazole and Salicylic acid obtained by analyst 1 and analyst 2. The results are illustrated in (Table 8). The result showed that the % RSD was less than 2%.

Table 8: Result of Ruggedness of Ketoconazole and Salicylic acid for Method I and Method II.

Method	Drugs	Conc (µg/ml)	Wavelength (nm)	Analyst 1		Analyst 2	
				Absorbance* mean ± SD	%RSD	Absorbance* mean ± SD	%RSD
Method I	Keto	15	225	0.6643 ± 0.0035	0.52	0.668 ± 0.003	0.44
	SA	15	296	0.3883 ± 0.0030	0.78	0.3916 ± 0.0025	0.64
Method II	Keto	15	239	0.4477 ± 0.0015	0.34	0.4517 ± 0.0020	0.46
			296	0.055 ± 0.001	1.81	0.057 ± 0.001	1.75
	SA	15	239	0.472 ± 0.003	0.63	0.471 ± 0.002	0.53
			296	0.3883 ± 0.0030	0.78	0.3917 ± 0.0025	0.64

Application of developed UV spectrophotometric methods

The developed methods were applied successfully to the emulgel formulation. The Percentage of Ketoconazole and Salicylic acid from the emulgel dosage form was found to be 101.66% and 90.53% by Simultaneous equation method (Method I) and 98.92% and 91.61% by Q-absorbance ratio method (Method II) respectively with standard deviation less than 2%.

DISCUSSION

Method I rely on resolution of a simultaneous equation. Absorbances of Ketoconazole and Salicylic acid in ethanol and buffer pH-5 in ratio 1:1 solvent was determined at their respective absorbance maxima's (λ_{max}) of 225 and 296 nm. Method II is the Q-analysis or absorption ratio method in which absorbances were determined at 239 nm (isosbestic point) and 296 nm (λ_{max} of Salicylic acid). The methods are validated in accordance with the

International Conference on Harmonization (ICH) requirements. The proposed method's reproducibility, repeatability, and accuracy were determined to be satisfactory, as evidenced by the low standard deviation (SD) and % relative standard deviation values (%RSD). Recovery studies, in which known amount of the drugs were added to the reanalyzed formulations have explored the validity and reliability of the proposed methods. The % recovery attained specifies that the additives used in the emulgel formulation do not interfere. The % RSD value calculated from the robustness study was found to be less than 2% for both drugs indicating that the method is robust. Thus, the developed methods, simultaneous equation method (Method I) and Q-Absorbance Ratio Method (Method II) in current study is found to be accurate, precise, sensitive and simple. The assay results indicates that these methods can be effectively used for simultaneous estimation of both drugs in formulation. The proposed methods use inexpensive laboratory reagents, solvents and tools. As a result, these procedures can be easily implemented in quality control laboratories for routine analysis.

CONCLUSION

Ketoconazole and Salicylic acid are known for its wide therapeutic application and hence can be formulated into a series of pharmaceutical dosage forms. Two simple, precise and economic Spectrophotometric methods such as Simultaneous estimation method and Absorbance ratio method were developed for simultaneous estimation of both the drug Ketoconazole and Salicylic acid in bulk and in developed emulgel. This method can be extended as a challenging approach to formulate dosage forms containing Ketoconazole and Salicylic acid in combination. The developed analytical method can be used to perform routine analysis and quality control on pharmaceutical formulations that contain these drugs in combination.

CONFLICT OF INTEREST

Authors declare that there is no conflict of interest.

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