IJCRT.ORG ISSN : 2320-2882

INTERNATIONAL JOURNAL OF CREATIVE RESEARCH THOUGHTS (IJCRT)

An International Open Access, Peer-reviewed, Refereed Journal

METHOD DEVELOPMENT AND VALIDATION FOR ESTIMATION OF TRIAMCINOLONE IN TABLET AND INJECTION DOSAGE FORMS BY UV SPECTROPHOTOMETRY

Abirami G[1], Sanjeev Kumar V[2], Sivasanmugasundaram S[2], Venkadesan R. K[2]

[1]Associate Professor, [2]Students

Department of Pharmaceutical Analysis,

Adhiparasakthi College of Pharmacy, Melmaruvathur-603319, Tamilnadu,

affiliated to The Tamilnadu Dr. M.G.R. Medical University, Chennai-600032, Tamilnadu, India.

Abstract: Triamcinolone (TC) determination in bulk and tablet and injection dosage forms has been made possible by the development of sensitive, quick, economical and accurate UV spectrophotometric approach. The 242 nm wavelength was used for quantification. Beer's law was followed at concentrations between 2 to 12 µg/ml. ICH guidelines were followed in the method's validation. A statistical analysis demonstrated that the technique was reliable for the analysis of TC in tablet and injection dosage forms. The proposed technique was shown to be suitable for routine analysis and quality control assay of tablet and injection due to its wide linearity, range, sensitivity, accuracy, and simple procedure.

Index Terms **- Triamcinolone, Raw Material, UV Method, Tablet and Injection Dosage Forms.**

I. INTRODUCTION

Triamcinolone is a synthetic corticosteroid drug that belongs to the glucocorticoid class of drugs. It is used as anti-inflammatory and immunosuppressive agent. Molecular weight is 434.5 g/mol. Molecular formula is $C_{24}H_{31}FO_6$. IUPAC name is (11β,16α)-9-Fluoro-11,16,17,21-tetrahydroxypregna-1,4-diene-3,20-dione[1,2,3,4]. The literature reported few analysis methods for quantification of Triamcinolone in topical dosage forms only. However, no analytical method has been developed for determination of Triamcinolone in tablet and injection formulation by UV Spectroscopic method. Therefore, the aim of this work is to develop a simple, accurate and reliable UV Spectroscopic method for the Triamcinolone in tablet and injection dosage forms determination. ICH guidelines were followed in the method's validation[5&6].

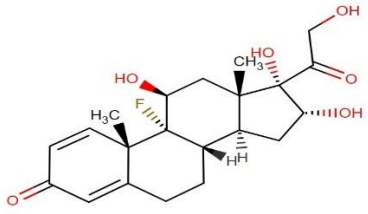

Figure 1: Structure of Triamcinolone

II. MATERIALS AND METHODS:

Instrumentation:

Shimadzu double beam UV/Visible spectrophotometer (Model UV-1700) with spectral band width of 1 nm instrument was utilized. The Shimadzu AUX-220 electronic balance was used for weighing all the samples.

Reagents and chemicals:

The pharmaceutical dosage forms used in this study was a KENACORT tablet and KENACORT® Injection I.P manufactured by Abbott Health Care LTD. (Mumbai) labelled to contain 4mg/tab and 40mg/ml of Triamcinolone. Methanol and distilled water was used as solvents.

Selection of solvent:

An important step in the development of a method is the selection of solvent for UV analysis. For the quantification of Triamcinolone for the UV Spectrophotometric technique, methanol and distilled water was the solvent utilized. In most reported methods alcoholic solvents were used, so in this method Hydro-alcoholic solvent system was used.

Preparation of standard stock solution:

Standard stock solution of Triamcinolone (100µg/ml) was prepared by dissolving 25 mg of Triamcinolone in methanol and distilled water in 100 ml volumetric flask.

Selection of wavelengths for estimation:

In this UV spectrophotometric method, solution of Triamcinolone (10 µg/ml), was prepared by appropriate dilution of standard stock solution with distilled water and scanned in the spectrum mode from 200nm to 400 nm. From the absorption spectra obtained, the wavelength selected for quantification was 242 nm.

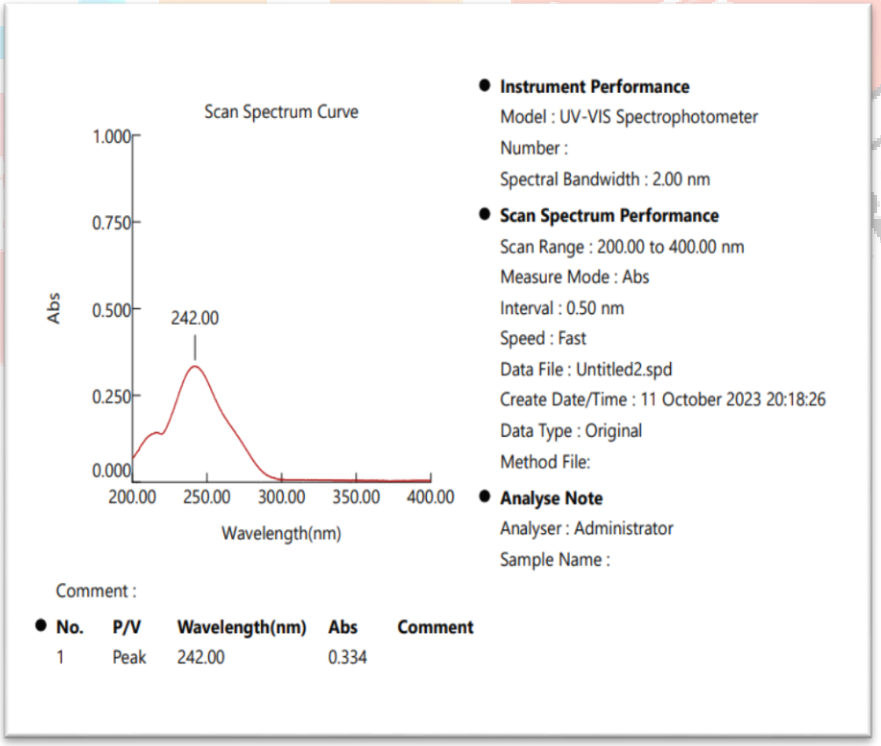

Figure 2: UV Spectrum of Triamcinolone

Preparation of calibration graph:

From the working standard solution of Triamcinolone containing 20µg/ml pipetted out 1, 2, 3, 4, 5 and 6 ml was transferred into series of six 10 ml volumetric flasks and made up to the volume with distilled water to get the final concentration range of 2, 4, 6, 8, 10 and 12µg/ml. The absorbances of different concentration solutions were measured at 242nm.

Quantification of Formulation (Tablet):

The average weight of twenty tablets, each of which contained 4 mg of TC, was estimated and the tablets were then crushed to get a fine powder. A quantity of powder equal to 25 mg of TC was transferred to a clean 100 ml volumetric flask. Methanol and distilled water was added to dissolve and the mixture was sonicated for 20 minutes. Subsequently, more distilled water was added to made upto the mark. Filtered the solution and the filtrate was suitably diluted to get the final concentration 4µg/ml was measured the absorbance at 242 nm using distilled water as blank. The concentration of TC was determined. Calibration curve were used to determine the concentration of TC in the diluted solution. Following that, the amount of TC in mg/tab was determined by multiplying the measured concentration by the dilution factor. The absorbance measurements were made six times for formulation and shown in TABLE-3.

Quantification of Formulation (Injection):

1 ml of formulation equivalent to 40 mg was pipetted out and transferred to 100ml volumetric flask, added minimum amount of methanol, distilled water to dissolve and made-up to the volume with distilled water to get final concentration of 4 µg/ml. The absorbance measurements were made six times for formulation at 242nm using distilled water as blank. The concentration of TC was determined and the amount of Triamcinolone present in formulation was calculated from the slope and intercept of respective calibration curve and shown in TABLE-4.

Validation:

The developed method for Triamcinolone was validated as per ICH norms.

Accuracy:

Accuracy of the developed method was confirmed by recovery analysis. Recovery studies were conducted using the standard addition method at three different levels (80%, 100%, and 120%) to evaluate the recovery studies of the suggested approaches for both the tablet and injection dosage forms. The recovery study findings, shown in TABLE – 05 & 06 as a percent recovery were good.

Precision:

The reproducibility of this method was determined by analysing both tablet and Injection (Intra-day assay precision) at different time intervals on same day for four times and (Inter-day assay precision) on four different days. Coefficient of variance for inter-day assay precision was found to be 0.4592 for TC and Intra-day assay precision was found to be 0.4569 for TC in tablet dosage form. Similarly, the Coefficient of variance for inter-day assay precision was found to be 0.3649 for TC and Intra-day assay precision was found to be 0.5982 for TC in Injection formulation.

III. RESULTS AND DISCUSSION

The UV Spectroscopic Method has advantages that it takes less time and consume less solvent. To ensure the % purity in two different dosage forms of the drug, the UV-spectroscopy was developed.

An accurate, fast, simple and precise UV Spectrophotometric method was developed and validated. Methanol and distilled water was chosen as a solvent for the estimation of TC. The standard solution of 10 µg/ml of TC in the appropriate solvents were made, and the solution was scanned with distilled water in the UV region between 200 and 400 nm to record the spectra, which is shown in Figure- 02. 242 nm was selected from the UV spectrum to determine the TC. The aliquots of six different TC concentrations ranging from (2.0 to 12.0 g/ml) were prepared. For the UV spectrum, the absorbances were measured at 242 nm. For the drug at 242 nm wavelength, the calibration graph preparation process was done six times, calculated and shown in TABLE-01. On the calibration graph, absorbances were plotted using absorbance against concentration. For the TC drug, optical parameters like the correlation coefficient, Sandell's sensitivity, LOQ, LOD, standard error and molar absorptivity were calculated and shown in TABLE-02. It was found that TC correlation coefficient was found to be 0.9996. Thus, it was showen that the calibration graphs were linear. Kenacort tablet formulation containing 4 mg & Kenacort Injection- 40mg/ml of TC was selected for estimation.

From the linearity, the nominal concentration of TC i.e. 4 µg/ml was prepared. Six test solution were determined based on the absorbance of the solutions was measured at 242 nm. The % purity found in tablet dosage form was 98.125 ± 0.3604 and injection dosage form was 99.45 ± 0.4057. The % RSD values were found to be very less. Hence the method has good precision. The results of analysis are shown in TABLE-03&04. The precision of the developed method was confirmed by repeated analysis of the formulation for six

times with the same concentration (Intraday and Interday). The % RSD values for Intraday and Interday analysis were found to be 0.4569 & 0.4592 for the tablet and 0.5982 & 0.3649 for the injection dosage forms and shown in TABLE- 09 & 10. The results show the developed UV method was very high. In ruggedness studies, Different Instrument 1, 2 and 3 were found to have % RSD values of 0.9933, 1.2827 and 0.9378 respectively. Different Analyst 1, 2 and 3 were found to have % RSD values of 0.3585, 0.5553 and 1.3337 respectively for the tablet dosage form. In the same way the studies in injection dosage form were conducted and Different Instrument 1, 2 and 3 were found to have % RSD values of 0.5635, 0.9398 and 1.2624 respectively. Different Analyst 1, 2 and 3 were found to have % RSD values of 0.5559, 1.3328 and 0.3597 respectively and shown in TABLE – 07 & 08. Lower % RSD values indicated, method was more rugged. Accuracy of the developed UV method was confirmed by recovery analysis. The % recovery ranged from 100.25-102.29% for tablet and 101.60-102.00% for injection. Lower % RSD values indicated that the developed UV spectroscopic method was more accurate. The results was shown in TABLE - 05 & 06.

Table 01: Average of Linearity Values

S.No	Concentration (µg/ml)	Absorbance
1.	2	0.083
2.	4	0.154
3.	6	0.236
4.	8	0.303
5.	10	0.376
6.	12	0.444

Table 02: Optical Characteristics of Triamcinolone

S.NO	PARAMETER	AT 242 nm*
1	Molar absorptivity (L $mol^{-1}cm^{-1}$)	16342.37
2	Regression Equation (y= mx+c)	y = 0.0369x+0.0066
3	Slope (m)	0.0369
4	Intercept (c)	0.0066
5	Correlation coefficient (r^2)	0.09996
6	Sandell's sensitivity (µg/cm^2/0.001 A.U)	0.0270
7	LOD (µg/ml)	0.0961
8	LOQ (µg/ml)	0.2914

*Mean of six observations

Table 03: Quantification of Formulation (Kenacort Tablet)

S.NO	Labelled Amount (mg/tab)	Amount Found (mg/tab)*	Percentage Obtained (%w/w)*	Average of % found (%w/w)	S.D	% R.S.D
1	4	3.96	99.00			
2	4	3.90	97.25			
3	4	3.91	97.5	98.125	0.8829	0.8982
4	4	3.915	97.75			
5	4	3.99	99.75			
6	4	3.94	98.5			

*Mean of six observations

Table-04: Quantification of Formulation (Kenacort® Injection)

S.NO	Labelled Amount (mg/ml)	Amount Found (mg/ml)*	Percentage Obtained (%w/v)*	Average of % found (%w/v)	S.D	% R.S.D
1	40	39.8	99.50			
2	40	39.3	98.25			
3	40	40.01	100.25	99.45	0.9939	0.9993
4	40	39.8	99.5			
5	40	40.04	101.00			
6	40	39.3	98.25			

*Mean of six observations

Table- 05: Recovery Study (Kenacort Tablet)

Sample	% concentration	Sample Amount* (µg/ml)	Amount spiked* (µg/ml)	Estimated Amount* (µg/ml)	Recovered Amount* (µg/ml)	Average % Recovery*	S.D	%R.S.D
KNC TAB	80	4	3.2	7.28	3.28	100.95	0.8463	0.8366
	100	4	4	8.07	4.07	100.25		
	120	4	4.8	8.91	4.91	102.29		

*Mean of six observations

Table- 06: Recovery Study (Kenacort Injection)

Sample	% concentration	Sample Amount* (µg/ml)	Amount spiked* (µg/ml)	Estimated Amount* (µg/ml)	Recovered Amount* (µg/ml)	Average % Recovery	S.D	%R.S.D
KNC INJ	80	4	3.2	7.26	3.26	102.00	0.1639	0.1610
	100	4	4	8.12	4.12	101.60		
	120	4	4.8	8.88	4.88	101.77		

*Mean of six observations

Table-07: Ruggedness Study Formulation (Kenacort Tablet)

SAMPLE	TYPE OF RUGGEDNESS	AVERAGE* % RECOVERY	S.D	% R.S.D
TC	Analyst – 1	97.5	0.3494	0.3585
	Analyst – 2	99.5	0.5524	0.5553
	Analyst – 3	98.5	1.3132	1.3337
	Instrument- 1	100.26	0.9960	0.9933
	Instrument- 2	98.68	1.2659	1.2827
	Instrument- 3	97.5	0.9244	0.9378

*Mean of six observations

Table- 08: Ruggedness Study Formulation (Kenacort Injection)

SAMPLE	TYPE OF RUGGEDNESS	AVERAGE* % RECOVERY	S.D	% R.S.D
TC	Analyst – 1	99.55	0.5535	0.5559
	Analyst – 2	98.50	1.3150	1.3328
	Analyst – 3	97.65	0.3501	0.3597
	Instrument- 1	99.65	0.5610	0.5635
	Instrument- 2	97.5	0.9255	0.9398
	Instrument- 3	98.25	1.2403	1.2624

*Mean of six observations

Table- 09: Intraday and Interday Analysis of Kenacort Tablet

Sample	Sample number	Label claim (mg/ml)	% Purity* (% w/v)		SD		% RSD	
			Intraday	Interday	Intraday	Interday	Intraday	Interday
KNC INJ	1	40	98.75	98.18	0.5921	0.3601	0.5982	0.3649
	2	40	99.48	99.04				
	3	40	98.12	98.78				
	4	40	99.58	99.5				
Mean purity (% w/w)			98.98	98.66				

*Mean of Four observations

Table- 10: Intraday and Interday Analysis of Kenacort Injection

Sample	Sample number	Label claim (mg/tab)	% Purity* (% w/w)		SD		% RSD	
			Intraday	Interday	Intraday	Interday	Intraday	Interday
KNC TAB	1	4	99.5	99.59	0.4524	0.4549	0.4569	0.4592
	2	4	98.65	99.12				
	3	4	98.48	98.48				
	4	4	99.42	99.08				
Mean purity (% w/w)			99.012	99.06				

IV. CONCLUSION:

The validated spectrophotometric method employed here proved to be simple, economical, rapid, precise and accurate. Thus, these can be used for routine estimation of Triamcinolone in tablet & injection dosage forms.

REFERENCES

[1] Anonyms. https://go.drugbank.com/drugs/DB00620

[2] Anonyms. https://psiberg.com/uv-vis-spectroscopy/#google_vignette

[3] Anonyms. https://www.drugs.com/triamcinolone.html

[4] Anonyms. https://www.ncbi.nlm.nih.gov/books/NBK544309/

[5] International Conference on Harmonization (ICH) of Technical Requirements for the Registration of Pharmaceuticals for Human Use. Validation of Analytical Procedures, ICH-Q2A, Geneva; 1995.

[6] International Conference on Harmonization (ICH) of Technical Requirements for the Registration of Pharmaceuticals for Human Use. Validation of Analytical Procedures: Methodology, ICH-Q2B, Geneva; 1996.