ISSN No:-2456-2165 https://doi.org/10.38124/ijisrt/IJISRT24AUG262

Development of Spectrophotometric Method for the Quantitative Determination of Betamethasone 17-Valerate in Creams

Motiki F. Beleme[1]; Lebohang A. Moetseloa[2]; Letuka J. Sello[3]
Department of Research and Development, Tripharm Manufacturing Pty Ltd, Lesotho

Abstract:- A simple spectrophotometric technique has been developed and validated for determination of betamethasone 17-valerate in pharmaceutical formulations. The method is based on cream dissolution in absolute ethanol and assaying spectrophotometrically at 240nm. The amount of betamethasone 17-valerate in cream was determined as 101.6 ± 0.0037 % w/w. The method validation demonstrated linearity for concentration range of 0.006mg/ml to 0.0014mg/ml (R^2=0.9995). The method is simple and accurate and has successfully been employed in the analysis of some of Tripharm pharmaceutical formulations.

Keyword:- *Betamethasone 17-Valerate, Spectrophotometry, Betamethasone, HPLC.*

I. INTRODUCTION

The chemical designation of betamethasone 17-valerate (BV) is (8S,9R,10S,11S,13S,14S,16S,17R)-9-fluoro-11-hydroxy-17-(2-hydroxyacetyl)-10,13,16-trimethyl -3-oxo-6,7,8,9,10,11,12,13,14,15,16,17-dodecahydro-3H-cyclopenta[a]phenanthren-17-yl pentanoate (da Silva Solon *et al.*, 2016). This strong corticosteroid, which is stated as betamethasone base, comes in cream, ointment, lotion, or gel forms and has a 0.1% (w/w) strength (Smith, Haigh and Kanfer, 1985). Betamethasone 17-valerate is a white powder that is glucocorticoid-active and semi-synthetic pharmaceutical raw material that dissolves slowly in water and ethanol (Permata *et al.*, 2019). Synthetic corticosteroids like betamethasone 17-valerate are frequently used in dermatological preparations, especially creams, to treat a range of skin disorders such dermatitis, psoriasis, and eczema. Additionally, BV is frequently found in dermatological formulations that are used to treat a range of skin disorders, including inflammation (Smith, Haigh and Kanfer, 1985; Deceuninck *et al.*, 2011).

For quality control and therapeutic efficacy, it is imperative that the concentration of betamethasone 17-valerate in these creams be determined precisely. To assure the safety and efficacy of topical dermatological medicinal products, quality control requires extensive and high-level analytical support (Smith, Haigh and Kanfer, 1985). Distinct liquid chromatographic techniques have been documented for the assessment of betamethasone 17-valerate either independently or in conjunction with other ingredients in various formulations (USP 41, NF36 (2018); BP2018).

The determination of betamethasone 17-valerate in pharmaceutical preparations has been studied using a variety of analytical techniques in the past. These techniques include spectrophotometry, high-performance liquid chromatography (HPLC), and liquid chromatography-mass spectrometry (LC-MS). Chromatographic techniques provide a high degree of sensitivity and selectivity, but they can also be costly to use and take a while to analyze (Gupta *et al.*, 2022). On the other hand, spectrophotometry is a commonly used analytical technique that provides a dependable and affordable way to measure the medication in pharmaceutical formulations.

Low-cost, quick, and effective ways to test the quality of prescription goods are valuable because of the pharmaceutical industry' global expansion (Smith, Haigh and Kanfer, 1985). The available literature indicates that spectrophotometry holds promise for the quantitative examination of betamethasone 17-valerate (Awen *et al.*, 2010; Gupta *et al.*, 2022). The focus is on refining parameters including solvent choice, wavelength choice, and calibration curve creation. The goal of this work is to develop a cheaper, reliable and effective assay for pharmaceutical determination of betamethasone 17-valerate in creams using spectrophotometer.

II. EXPERIMENTAL

➢ *Material*

Betamethasone cream was produced by Tripharm manufacturing company using betamethasone 17-valerate which was sourced from the Kirsch Pharma South Africa. Betamethasone 17-valerate reference standard from laboratory of Dr. Ehrenstorfer Augsburg-Germany, 99.7% Ethanol HPLC grade from VWR Chemicals was used as a solvent, 10 mL screw cap syringes and 0.45µm membrane filters and the Shimadzu UV-1900 UV-VIS spectrophotometer and quartz cuvette of 10 mm path length were used in this study.

ISSN No:-2456-2165 https://doi.org/10.38124/ijisrt/IJISRT24AUG262

➢ *Standard Preparation*

A 1.00 mg/ml stock solution of betamethasone 17-valerate standard was prepared by dissolving 0.100 g of standard in absolute ethanol and diluted to 100 ml with the same solvent. A 1.0 ml of 1.00 mg/ml was pipetted in a glass beaker and diluted to 10 ml with absolute ethanol to make a 0.1 mg/ml solution which were serially diluted to make concentration of 0.006 mg/ml, 0.008 mg/ml, 0.010 mg/ml, 0.012 mg/ml and 0.014 mg/ml solutions. The standards were prepared using BV secondary standard after discovery that both primary and secondary standards have similar absorption spectrums.

➢ *Betamethasone Cream Sample Preparation*

A number of eight samples were prepared from a recently produced cream. For each sample a 0.1000 g of betamethasone 0.1% cream was dissolved in 10 ml of absolute ethanol and the warmed in a water bath to complete dissolution, cooled and left overnight. Each sample was filtered with a 0.45 um membrane filter the next day.

➢ *Spectrophotometric Analysis of Solutions*

The UV-Vis spectrophotometer was auto zeroed using absolute ethanol and each standard aliquot was added into the quartz cuvette and inserted into the cuvette holder and then analyzed at 240 nm. The betamethasone 0.1% cream filtrates were also analyzed in a same technique. The results for spectrophotometric analysis of standard solutions were used to generate multiple-point calibration curve which was used in the calculation sample concentration.

➢ *Determination of Betamethasone Maximum Absorbance Wavelength*

A 0.1000 g of Betamethasone 17-Valerate standard was dissolved in HPLC grade ethanol and diluted to 100 mL with the same solvent. The solution was then diluted by 2-fold dilution and then 10-fold dilution respectively. It was run at a range of 300 to 200 nm. The wavelength for analysis of samples was chosen at 240nm because the figure 1 below of betamethasone 17-velarate spectrum shows maximum absorption at 240nm.

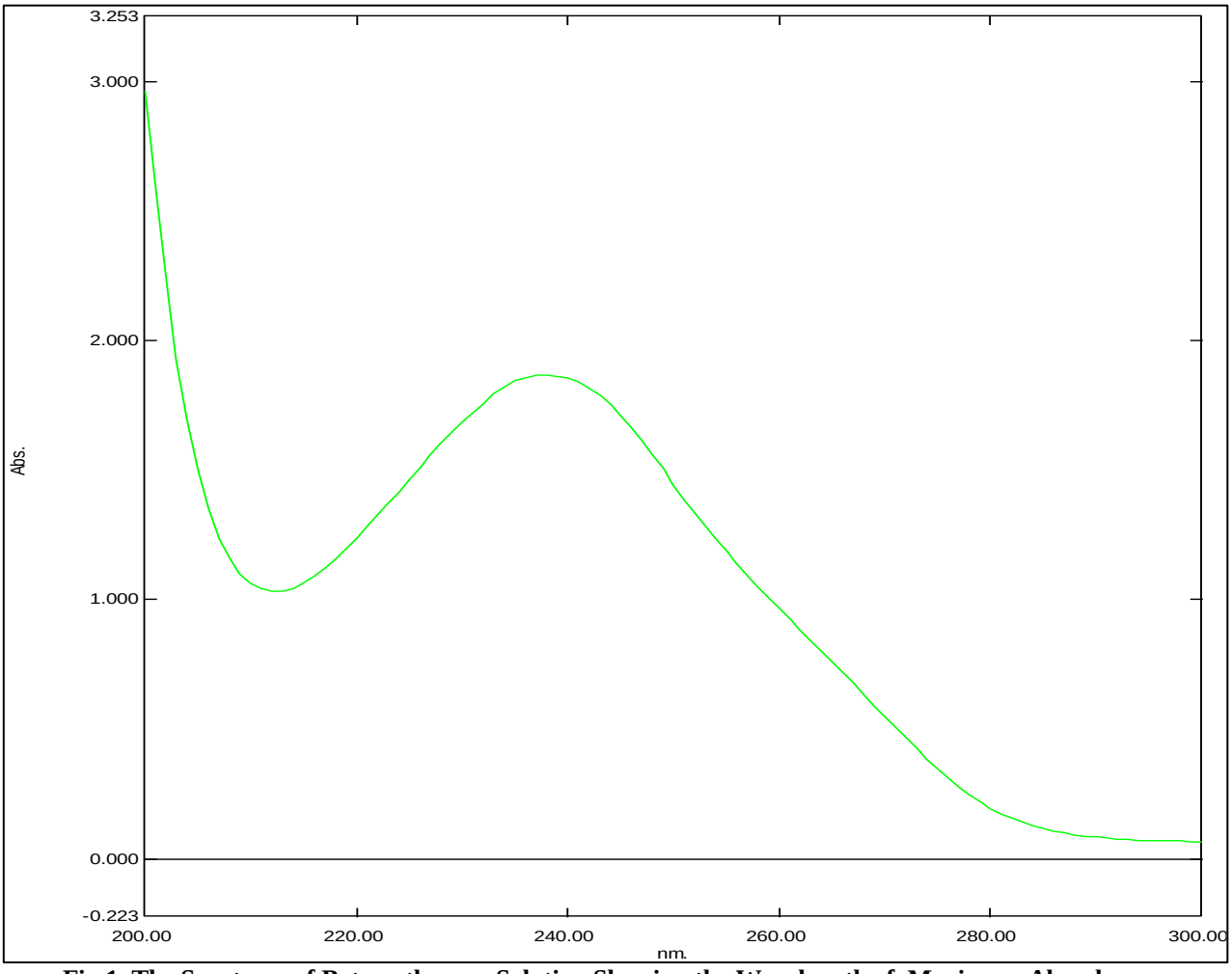

Fig 1. The Spectrum of Betamethasone Solution Showing the Wavelength of Maximum Absorbance.

Caution! Ethanol is highly flammable liquid and vapor keep away from heat and spark. Wear protective clothing.

ISSN No:-2456-2165 https://doi.org/10.38124/ijisrt/IJISRT24AUG262

III. RESULTS AND DISCUSSION

The proposed method was validated based on adapted ICH guidelines by (Walfish, 2006) for its linearity, precision, accuracy and limit of detection. The method offers simplicity and prompt analysis. Results for analysis of betamethasone cream as well as statistical parameter computed from calibration curve are tabulated in tables 1 and 2 below.

Table 1: Mass and Absorbance of Betamethasone Cream Samples at 240nm

Trial	Mass /g	Absorbance at 240 nm	Concentration mg/mL	%(w/w)
1	0.1232	0.501	0.01489	0.1208
2	0.0991	0.435	0.01290	0.1301
3	0.1081	0.433	0.01284	0.1188
4	0.1124	0.461	0.01368	0.1217
5	0.1110	0.444	0.01317	0.1186
6	0.1010	0.433	0.01284	0.1271
7	0.1041	0.449	0.01332	0.1279
8	0.1035	0.444	0.01317	0.1272

Table 2: Optical and Regression Characteristics of the Method

Parameter	Value
Λ(nm)	240
Beer's law limit(mg/ml)	0.006-0.014
Molar absorptivity(L/mL/cm)	1.606×10^{4}
LOD(mg/ml)	3.828×10^{-4}
Linear regression equation Y=a +bc	
Slope(b)	33.15
Intercept(a)	0.0075
Coefficient of determination(R^2)	0.9995

IV. METHOD VALIDATION

➢ *Accuracy*

The amount of betamethasone 17-valerate added during manufacturing of the cream which is 0.122 g and the amount found by spectrophotometric analysis were subjected to statistical analysis to validate the results and investigate whether the discrepancy between the two values arise from systematic errors. The performed t-test indicated that the results are affected by indeterminate sources of errors. The proposed analytical method provides results with satisfactory degree of accuracy as it yielded a small margin of error of 1.63% and percentage recovery of 101.6 ± 0.003786 % w/w which shows that other excipients do not interfere with the analysis of the cream.

➢ *Precision*

Eight samples were run on the same day under the same operating conditions using the same instrument. About three minutes' intervals were monitored for each sample to be injected into the instrument and the data was recorded. The relative standard deviation of 3.65% was calculated from the data revealed that the method has satisfactory precision as value of RSD of this intra-assay precision is less than 5%.

➢ *Linearity*

Different concentrations of Betamethasone 17-valerate standard solutions prepared from 1.0 mg/ml stock solution were used to determine the linearity of the method. The linearity of Betamethasone 17-valerate was found on the concentration range of 0.006 mg/ml to 0.014 mg/ml and resulted in coefficient of determination (R^2) valued at 0.9995.

➢ *Limit of Detection*

A low limit of detection indicates that the method can detect analyte at lower concentration hence, greater sensitivity. This spectrophotometric method for determination of betamethasone 17-valerate in cream has capability to detect analyte in trace level in complex matrix making it applicable for quantification of analyte at lower concentrations as low as 3.828×10^{-4} mg/ml.

➢ *Application of the Method*

The proposed method has been employed in the assay of betamethasone 0.1% cream and hydrocortisone 1% cream products. The results in both creams were in agreement with the amount of added active ingredients. The method is much easier to employ since it does not require critical reaction conditions, tedious sample preparation and the reagents are cheap and readily available.

ISSN No:-2456-2165 https://doi.org/10.38124/ijisrt/IJISRT24AUG262

V. CONCLUSION

The study has successfully achieved development of cheaper spectrophotometric method for quantitative determination of Betamethasone 17-valerate as the method demonstrated high efficiency and accuracy with acceptable percentage recovery and the lowest possible percentage error. This method facilitates production of high quality grade Betamethasone17-valerate containing pharmaceutical products by performing post-production quality control analysis which is tasked to ensure that the products meet the required standard. Therefore, the method can be used for routine analysis of betamethasone and hydrocortisone in pharmaceutical formulation.

- *Conflict of Interest*

The authors declare no conflict of interest in publication of this work.

ACKNOWLEDGEMENTS

The authors appreciate Tripharm manufacturing company management for approval to publish the paper.

- *Supporting Information*

Additional experimental and statistical data including calibration curve.

REFERENCES

[1]. Asra, R., Rivai, H. and Astuty, W. (2017) 'Pengembangan dan Validasi Metode Analisis Betametason Tablet dengan Metode Absorbansi dan Luas Daerah di Bawah Kurva Secara Spektrofotometri Ultraviolet', *Jurnal Farmasi Higea*, 9(2), pp. 118–126.

[2]. Awen, B. *et al.* (2010) '2. Development and validation of a new spectrophotometric method for the determination of betamethasone in bulk and pharmaceutical dosage forms', *International Journal of Pharmaceutical and Biomedical Research (IJPBR)* [Preprint].

[3]. Deceuninck, Y. *et al.* (2011) 'Determination of MRL regulated corticosteroids in liver from various species using ultra high performance liquid chromatography–tandem mass spectrometry (UHPLC)', *Analytica chimica acta*, 700(1–2), pp. 137–143.

[4]. Gupta, M.K. *et al.* (2022) 'A comparative review on High-Performance Liquid Chromatography (HPLC), Ultra Performance Liquid Chromatography (UPLC) & High-Performance Thin Layer Chromatography (HPTLC) with current updates', *Current Issues in Pharmacy and Medical Sciences* [Preprint].

[5]. Nikolin, B. *et al.* (2004) 'High perfomance liquid chromatography in pharmaceutical analyses', *Bosnian journal of basic medical sciences*, 4(2), p. 5.

[6]. Permata, Y. *et al.* (2019) 'Formulation and Quantitative Analysis of Betamethasone Valerate and Neomycin Sulfate Cream by High Performance Liquid Chromatography and Spectrophotometry', *Open Access Macedonian Journal of Medical Sciences*, 7, pp. 3841–3846. Available at: https://doi.org/10.3889/oamjms.2019.516.

[7]. da Silva Solon, L.G. *et al.* (2016) 'Development and validation of an UHPLC method for the determination of betamethasone valerate in cream, gel, ointment and lotion', *Steroids*, 106, pp. 70–77.

[8]. Smith, E.W., Haigh, J.M. and Kanfer, I. (1985) 'A stability-indicating HPLC assay with on-line clean-up for betamethasone 17-valerate in topical dosage forms', *International journal of pharmaceutics*, 27(2–3), pp. 185–192.

[9]. Walfish, S. (2006) 'Analytical methods: a statistical perspective on the ICH Q2A and Q2B guidelines for validation of analytical methods', *BioPharm International*, 19(12), pp. 1–6.