Asian Journal of Chemistry Vol. 22, No. 2 (2010), 1649-1651

NOTE

Spectrophotometric Method for Estimation of Leflunomide in Bulk and Tablets

N. Rabindra Pal, Manas Chakraborty, Rabindra Debnath and Bijan K. Gupta*
Calcutta Institute of Pharmaceutical Technology and Allied Health Sciences, Division of Pharmaceutics, Howrah-711 316, India
E-mail: pal.rabindranath@gmail.com

A simple and sensitive UV spectrophotometric method for determination of leflunomide in bulk and tablets has been developed. Beer' law is obeyed in the concentration of 8-30 mcg/mL of leflunomide. The method was found to be simple, precise, accurate and cheap for routine analysis with recovery of 99.06 ± 0.7593 %. The method does not require any separation of soluble excipient in the tablet as they do not interfere in the estimation. Results of analysis was verified statistically and by recovery studies thus the method can be used for routine, quality control of bulk and other formulations.

Key Words: UV spectrophotometry, Leflunomide.

Chemically leflunomide is 5-methyl-N-[4-(trifluromethyl)phenyl]isoxozole-carboxamide, an immunomodulator pro drug which is used in rheumatoid arthritis and psoriatic arthritic[1]. It is a disease modifying antirheumatic drug.

A survey of literature reveals that HPLC and gas chromatography methods[2-4] are available for estimation of leflunomide. But there is no evidence in the literature for estimation of this drug by UV spectrophotometric method which is essential for routine quality control analysis of pharmaceutical products of leflunomide as a fast, selective and economical method. So an attempt has been made to develop a simple rapid and reproducible UV spectrophtometric method with greater precision accuracy for analysis of leflunomide in bulk and tablet for analysis.

A double beam UV-vis spectrophotometer (UV- 1700 Shimadzu, Japan) was employed with spectral bandwidth of 1 nm and wavelength accuracy of ± 0.3 nm with a pair of 10 mm quartz cells. All weights were taken on an electronic balance (AX-200, Shimadzu, Japan). Pure leflunomide was obtained as gift sample from M/s Aventis Pharmaceuticals Limited, Mumbai. Tablets of brand Arava ® containing 10 mg of leflunomide was procured from a local pharmacy. The solvents used for the experiment were ethanol (AR Grade, Qualigens Fine Chemicals, Mumbai) and doubly distilled water.

Preparation of standard drug solution: The standard solutions of leflunomide were prepared by dissolving 10 mg of drug in sufficient quantity of ethanol: water mixture (1:4 v/v) to obtain a final concentration of 100 mcg/mL. This stock solution

was used to prepare further dilutions of standard solution. Aliquots of stock solution of leflunomide were transferred into a series of 25 mL volumetric flasks and volume was made up to the mark with ethanol:water (1:4 v/v) mixture to produce the concentration ranging from 5-50 mcg/mL. 260 nm was selected as λ_{max} from the UV absorption spectra obtained by scanning the pure drug solutions for measuring the absorbance of above solutions to prepare the calibration curve.

Preparation of calibration curve: The solution were scanned in the UV range of 200-400 nm against blank. The absorption maxima were found to be 260 nm. The calibration curve was plotted (Fig. 1) with the absorbance readings against the corresponding concentrations. The optical characteristics for this method are given in Table-1.

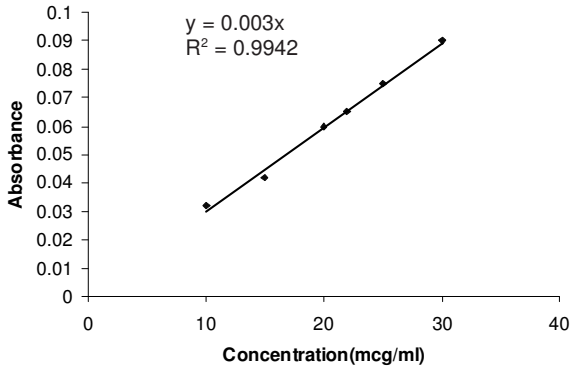

Fig. 1.

TABLE-1
OPTICAL CHARACTERISTIC AND PRECISION OF PROPOSED METHOD

Parameters	Values
Absorbance (nm)	260 nm
Beer's law limit (mcg/mL)	8–30
Corresponding coefficient (r)	0.9996
Molar absorptivity (l../mol/cm)	17197.6
Regression eqation (Y = mX + C)	Y = 0. 03253X + 0.00665
Slope (m)	0.03253
Intercept (C)	0.00665
Standard Deviation	0.30125
Precision (% RSD)	0.99321
% Range of Error: 95 % confidence limits	± 0.35464
99 % confidence limits	± 0.62656
Limit of detection (LOD)	0.28 (µg/mL)
Limit of Quantitation (LOQ)	0.835 (µg/mL)
% Recovery ± S.D.	99.06 ± 0.75

Analysis of marketed tablets: For the analysis of the dosage form, 20 tablets of leflunomide were ground to fine powder and mixed thoroughly. Tablet powder equivalent to 10 mg of leflunomide was transferred in about 40 mL of ethanol: water mixture (1:4 v/v) by shaking on a rotary flask shaker for 1 h. The solution was filtered through Whatman filter paper No. 41. The filter paper was washed with the blank. The washings were added to the filtrate and the final volume was made up to 100 mL with the blank. Suitable dilution was made so that the absorbance of the final sample comes corresponding to 30 mcg/mL against the blank at 260 nm. The amount of leflunomide was computed using the equation referring to the calibration curve. $Y = mX + C$ where C is concentration in µg/mL, Y is absorbance units. All readings are mean of six replicate samples. To examine the absorbance of either positive or negative interference of the excipients used in the formulation, recovery studies were carried out at five different levels by adding diluted pure drug solution equivalent to 0, 200, 400, 600 and 800 mcg of leflunomide to five samples of tablet powder solution containing equivalent amount of 1000 mcg of the drug. The determination was carried out for 5 replicates at each level.

The method was validated according to ICH guideline[5]. The optical characteristics such as Beer' law limits, molar absorptivity and other parameters in Table-1. The liner regression of absorbance on concentration gave the equation $Y = mX + C$ with a correlation coefficient (r) of 0. 99321 that indicates a good linearity between absorbance and concentration in the range of 8-30 mcg/mL. The value of percentage relative standard deviation less than 1 % and low percentage range of error confirm the high degree of precision and accuracy of the proposed method. The assay result obtained by the proposed method was found to be 99.9 ± 1.00011 % which is in good agreement with the label amounts. The % recovery value, which is closed to 100 %, indicates the reproducibility of the method and absorbance of interference of the excipients present in the tablets. The precision and accuracy were adequate over the concentration range 10 to 100 mcg/mL. These results show that the proposed method is suitable for its internal use.

Conclusion

The application of the proposed method has been performed in the pharmaceutical tablets of leflunomide and excellent result was found. The author conclude that the proposed spectrophotometric method for estimation of leflunomide is simple, sensitive, accurate, precise and reproducible and can be used for routine quality control analysis of leflunomide in bulk and in tablet formulations.

REFERENCES

1. Goodman and Gilman's, The Pharmacological Basis of Therapeutics, edn. 11, p. 1415 (2005).
2. D.S. Miron, C. Soldattelli and E.E.S. Schapoval, *Chromatographia*, **63**, 283 (2006).
3. D. Yeniceli, D. Dogrukol and M. Tuncel, *J. Pharm. Biomed. Anal*., **40**, 197 (2006).
4. P.V. Choudhari, T.S. Patil, N.A. Shah, M.H. Dehghan and A.G. Nikalge, *J. Pharm. Res.*, **7**, 68 (2008).
5. ICH Harmonized Taptrite Guideline, Recommended for Adoptation at Step 4 of the ICH Process on 6 November 1996 by the ICH Steering Committee.

(*Received*: 23 May 2009; *Accepted*: 5 November 2009) AJC-8034