Asian Journal of Chemistry Vol. 20, No. 1 (2008), 551-555

Reverse Phase HPLC Method for the Analysis of Terbinafine in Pharmaceutical Dosage Forms

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A rapid and sensitive high performance liquid chromatographic method was developed for the estimation of terbinafine in pharmaceutical dosage forms. Terbinafine was chromatographed on a reverse phase C_{18} column in a mobile phase containing buffer:acetonitrile in the ratio 65:35 v/v. The mobile phase was pumped at a flow rate of 1.8 mL/min and the eluents were monitored at 220 nm. The calibration curve was linear in the range of 20-1000 ng/mL. The intra- and inter-day variation was found to be less than 2 % showing high precision of the assay method. The mean recovery of the drug from the solution containing 20 ng/mL was 99.1 ± 0.73 % indicating high accuracy of the proposed HPLC method may be used for determining terbinafine in bulk drug samples or in pharmaceutical dosage forms.

Key Words: Dosage forms, Terbinafine, Reverse phase HPLC.

INTRODUCTION

Terbinafine is an antifungal agent and chemically is (E)-N-(6,6-dimethyl-2-hepten-4-ynyl)-N-methyl-1-naphthalene methanamine hydrochloride[1-5]. Several methods like voltammetry[6], electroanlytical methods[7], mass spectrometry[8] were reported for the estimation of terbinafine in pharmaceutical dosage forms. Some of the methods utilize liquid chromatography[9,10], gas chromatography[11] and these process are considered tedious. Other reported methods such as spectrophotometry[12,13] and HPLC[14-19] are not accurate and the process is considered tedious. The HPLC methods using the most commonly available columns and detectors like UV are preferred. The present study describes the determination of terbinafine in bulk drug samples and pharmaceutical dosage forms by using RP-C_{18} column with UV detector. Owing to the wide spread use of HPLC in routine analysis, it is important that HPLC methods are to be developed for estimating terbinafine. The aim of this study is to develop a simple, precise, rapid and accurate reversed phase HPLC method for the determination of terbinafine in bulk drug samples or in pharmaceutical dosage forms.

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EXPERIMENTAL

Terbinafine was gift sample from Novartis Pharmaceuticals Ltd, Worli, Maharastra, India. Acetonitrile and water were of HPLC grade (Qualigens). All other reagents (potassium dihydrogen phosphate) used in the study were of AR grade (Qualigens).

A isocratic HPLC (waters India, USA) with a single waters 510 pump, waters 486 tunable absorbance detector and RP-C_{18} column (Bondapak, 5 μm particle size) was used. The HPLC system was equipped with software Millennium 32.

Chromatographic conditions: The contents of the mobile phase, buffer (850 mg potassium dihydrogen phosphate in 1000 mL water, add 1 g of sodium 1-dec sulfonate and adjust the pH 3.0 with dilute phosphoric acid) and acetonitrile in the ratio 65:35 v/v were filtered before using through a 0.4 μm membrane filter and degassed for 0.5 h.

The components of the mobile phase were pumped from the solvent reservoir to the column at a flow rate of 1.8 mL/min that yielded column back pressure 140-150 kg/cm^2. The column temperature was maintained at 40 °C. The eluents were monitored at 220 nm. Prior to the injection of the drug solutions, the column was calibrated for at least 0.5 h with the mobile phase flowing through the systems.

Procedure: The solutions were prepared on a weight basis and volumetric flasks were used to minimize solvent evaporation. Stock solution of a drug was prepared by dissolving 100 mg of terbinafine in 100 mL volumetric flask containing 70 mL of methanol, sonicated for about 20 min and then made up to volume with mobile phase working standard solution of terbinafine was prepared by suitable dilution of the stock solution with mobile phase.

Five sets of the terbinafine were prepared in mobile phase at concentrations of 20, 40, 60, 80, 100, 200, 400, 600, 800, 1000 ng/mL. Each of this samples (20 μL) was injected five times into the column and the peak area of the drug was recorded.

Assay of terbinafine in tablets: 20 Tablets were weighed, finely powered and an accurately weighed sample of powdered tablets equivalent to 100 mg of terbinafine was placed in a 100 mL volumetric flask. 70 mL of methanol was added, shaken well and the flasks were allowed to stand for 4 h with intermittent sonication to ensure complete solubility of drug. The mixture was then made up to volume with mobile phase, thoroughly mixed and then filtered through a 0.4 μm membrane filter. An aliquot of the filtrate was transferred to a volumetric flask and made up to volume with mobile phase to give an expected concentration of 20 ng/mL of terbinafine. All determinations were conducted in triplicate.

Precision: The precision of the assay was determined in terms of intra- and inter-day variations in the peak area for a set of drug solution on three different days (n = 5). The intra- and inter-day variation in the peak area of drug solution(10 or 20 ng/mL)was calculated in terms of coefficient of variation (CV) and obtaining by multiplying the ratio of standard deviation to the mean with 100[CV= ± SD/mean × 100].

Accuracy: The accuracy of the HPLC assay method was assessed by adding known amount (10 or 20 ng) of the drug to a drug solution of known concentration (20 ng/mL) and subjecting the samples to the proposed HPLC method. The known amount of drug solution (10 or 20 ng/mL) was also added to the volumetric flask containing the powder sample of the tablet formulation with known amount of drug. The drug was estimated as the procedure described above for the estimation of terbinafine in the tablet formulations. In both the cases, the recovery studies were replicated five times. The accuracy was expressed in terms of the recovery and calculated by multiplying the ratio of measured drug concentration to the expected drug concentration with 100, so as to give the per cent recovery.

RESULTS AND DISCUSSION

The run time of the method was set at 35 min and terbinafine appeared on the chromatogram at 14.95 min. When the same drug solution was injected 5 times, the retention time of the drugs was same. The peak areas of terbinafine was calculated and the average of five such determinations were given in Table-1. When the concentration of terbinafine and its respective peak area were subjected to regression analysis by least square method, a high correlation coefficient was observed (r = 0.9999) in the range of 20-1000 ng/mL only. The regression of terbinafine concentration over its peak area was found to be Y = -339.07 + 142.73X where 'Y' is the peak area and 'X' is the concentration of terbinafine. This regression was used to estimate the amount of terbinafine either in tablet formulation or in validation study.

Proposed HPLC methods were also validated for intra- and inter-day variation. when the solutions containing 10 or 20 ng/mL of terbinafine were repeatedly injected on the same day, the coefficient of variation (CV) in the peak area of the drug for five replicate injection was found to less than 2 %. Also, the inter -day variation (3 and 5 injections) was found to be less than 2 % (Table-2). Thus the results have shown that the proposed HPLC method is highly reproducible. when a known amount of drug solution (10 or 20 ng) was added to a known concentration of drug solution (20 ng/mL), this was a high recovery (99.1 ± 0.73 %) of terbinafine (Table-3) indicating that the proposed method is highly accurate.

TABLE-1
CALIBRATION OF THE HPLC METHOD FOR THE ESTIMATION OF TERBINAFINE

Concentration of terbinafine (µg/mL)	Peak area	CV (%)
20	286.75	1.21
40	5791.50	1.83
60	8374.50	1.92
80	11406.40	2.10
100	13357.50	1.65
200	27856.10	0.41
400	55702.20	1.81
600	85842.30	0.62
800	113980.30	1.31
1000	142453.82	0.93

Regression equation (from 20 to 1000 µg/mL):
Y = -339.07 + 142.73X (r = 0.9999)

TABLE-2
PRECISION OF THE PROPOSED HPLC METHOD

Terbinafine concentration (ng/mL)	Concentration of terbinafine (ng/mL) found on			
	Inter-day		Intra-day	
	Mean (n = 5)	CV (%)	Mean (n = 5)	CV (%)
10	9.32	0.92	10.02	1.34
20	21.41	1.32	19.83	1.98

TABLE-3
RECOVERY OF TERBINAFINE

Amount of drug added (ng)	Mean (± SD) amount (ng) recovered (n = 5)	Mean (± SD) % of recovery (n = 5)
10	9.41 ± 0.32	94.1 ± 0.83
20	19.82 ± 0.31	99.1 ± 0.73

The HPLC method, developed in the present study has also been used to quantify terbinafine in tablet dosage forms. Terbinafine tablets (containing 250 mg of the drug) were analyzed as per the procedure described above. The average drug content was found to be 99 % of the labeled amount (Table-4). No interfering peaks were found in the chromatogram indicating that excipients used in the tablet formulations did not interfere with the estimation of drug by the proposed HPLC method.

TABLE-4
MEAN (± SD) AMOUNT OF TERBINAFINE IN TABLET DOSAGE FORMS BY PROPOSED HPLC METHOD

Brand of the tablet	Labeled amount (mg)	Observed amount (mg)	Purity (%)
AAA	250	248.3 ± 0.13	99.32 ± 0.92
BBB	250	246.8 ± 0.09	99.72 ± 1.12
CCC	250	248.8 ± 0.03	99.52 ± 0.82

ACKNOWLEDGEMENTS

The authors are thankful to the Management of College of Pharmaceutical Sciences, Mohuda, for providing facilities for developing HPLC method. The authors also thanks to M/s. Novartis Pharma Ltd., Worli, India for providing gift samples of terbinafine.

REFERENCES

1. The Merck Index, Merck & Co. Inc., New York, edn. 13, p. 937 (2001).
2. Wilson and Gisvold's, Text Book of Organic Medicinal and Pharmaceutical Chemistry, edn. 10.
3. P. Rodgers and M. Bassler, *Am. Family Physic.*, **63**, 663 (2001).
4. A.K. Gupta, N. Konnikov and C.W. Lynde, *J. Am. Acad. Dermatol.*, **44**, 479 (2001).
5. U.S. Food and Drug Administration (2001).
6. A. Arranz, S. Fernandez de Betono, J.M. Moreda, A. Cid, J.F. Arranz, *Anal. Chim. Acta*, **351**, 97 (1997).
7. S.A. Ozkan, B. Uslu, H.Y. Aboul-Enein, *Crit. Rev. Anal. Chem.*, **33**, 155 (2003).
8. C.H. de Olivera, R.E. Barrientos-Astigarraga, M.O. de Moraes, F.A.F. Bezerra, M.E.A. de Morraes and G. de Nucci, *Therap. Drug Monit.*, **23**,709 (2001).
9. T.K. Majumdar, R. Bakhtiar, D. Melaled and F.L.S. Tse, *Rapid Comm. Mass Spectr.*, **14**, 1214 (2000).
10. N. Brignol, R. Bakhtiar, L. Dou, T.K. Majumdar and F.L.S. Tse, *Rapid Comm. Mass Spectr.*, **14**, 141 (2000).
11. J. Kuzner, N.K. Erzen and M Drobnic-Kosorok, *Biomed. Chromatogr.*, **15**, 497 (2001).
12. Y.S. El-Saharty, N.Y. Hassan and F.H. Metwally, *J. Pharm. Biomed. Anal.*, **28**, 569 (2002).
13. S.G. Cardoso and E.E.S. Schapoval, *J. AOAC Int.*, **82**, 830 (1999).
14. M.H. Yeganeh and A.J. McLachlan, *Biomed. Chromatogr.*, **14**, 261 (2000).
15. S.G. Cardoso and E.E.S. Schapoval, *J. Pharm. Biomed. Anal.*, **19**, 809 (1999).
16. X.F. Ji, K.Y. Yu and W. Zhang, *Yaowu Fenxi Zazhi*, **17**, 363 (1997).
17. H. Zehender, J. Denoul, M. Roy, L. Le Saux and P. Schaub, *J. Chromatogr. B, Biomed. Appl.*, **664**, 347 (1995).
18. J. Denouel, H.P. Keller, P. Schaub, C. Delaborde and H. Humbert, *J. Chromatogr. B, Biomed. Appl.*, **663**, 353 (1995).
19. F. Schatz and H. Haberl, *Arzneim-Forsch.*, **39**, 527 (1989).

(*Received*: 30 November 2006; *Accepted*: 14 September 2007) AJC-5859