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Simple, Sensitive and Stability Indicating High Performance Liquid Chromatographic Assay of Terbinafine Hydrochloride in Dosage forms

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ABSTRACT

Terbinafine hydrochloride (TFH), is a potent antifungal agent of the allylamine class with broad spectrum activity against yeasts, dimorphic fungi, molds, and dermatophytes. A new, simple, rapid, selective, precise, accurate, and stability indicating high performance liquid chromatographic method has been developed for the determination of terbinafine hydrochloride (TFH) in pharmaceuticals. The assay was performed using an Zorbax Eclips XDB C-18 (3.5 µm, 4.6 × 150 mm i.d.,) column at 30ºC temperature with UV-detection at 222 nm. A mobile phase consisting of buffer (1000 mL water, 2 mL triethylamine, pH 3.4 adjusted with trifluoroacetic acid.), isopropyl alcohol and methanol (40:12:48, v/v/v), was used in the assay at a flow rate of 1 mL min^{-1}. The method was validated and system suitability parameters were investigated. An excellent linearity was obtained over the concentration range 1 - 80 µg mL^{-1} TFH with limits of detection (LOD) and quantification (LOQ) values of 0.3 and 1.0 mg mL^{-1}, respectively. The proposed method were applied successfully to the determination of TFH in tablets. The results obtained were in good agreement with those obtained by a reference method, with high precision and without any detectable interference from tablets excipients. The validity and reliability of the proposed methods were further assessed by the recovery studies *via* a standard addition method. In addition, forced degradation of TFH was conducted in accordance with the ICH guidelines. Acidic, basic water hydrolysis, thermal stress, peroxide and photolytic degradation were used to assess the stability indicating power of the method. Slight degradation was observed during oxidative degradation and no degradation was observed under other stress conditions.

Keywords: Terbinafine Hydrochloride, High performance liquid chromatography, Stability indicating, Pharmaceuticals.

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INTRODUCTION

Terbinafine hydrochloride (TFH), chemically known as (E)-N-(6,6-dimethyl- 2 - hepten - 4 - ynyl) -N-methyl - 1 – naphthalene methanamine hydrochloride [1], is a potent antifungal agent of the allylamine class with broad spectrum activity against yeasts, dimorphic fungi, molds, and dermatophytes [2-5].

Various techniques have been used for the determination of TFH in body fluids and pharmaceuticals. The drug is official in European Pharmacopoeia [6], British Pharmacopoeia [7] and the United States Pharmacopoeia [8]. European Pharmacopoeia and British Pharmacopoeia describe acid-base titration in hydro-alcoholic medium, the end point being located potentiometrically[6,7].United States Pharmacopoeia describes high performance liquid chromatographic method for the estimation of TFH[8]. High performance thin layer chromatography (HPTLC) has recently been applied for the assay of drug in tablets [9] and for the simultaneous determination of TFH and triamcinolone acetamide in compound tablets [10]. Non-aqueous titrimetry [11] visible spectrophotometry [12-14] and voltammetry [15] are the other analytical techniques available for the assay of TFH in its dosage forms. High performance liquid chromatography (HPLC) has been applied for the determination of TFH and metabolites in human plasma [16], human plasma, milk and urine[17], human plasma and urine [18] and the drug in tno.s [19]. An improved high throughput liquid chromatographic/tandem mass spectrophotometric method for TFH in plasma[20] and ultra performance liquid chromatographic method for the drug and its metabolites in human plasma and urine[21] are the other chromatographic methods reported for body fluids. In addition microbiological assay for the drug are also found in the literature[22-23].

HPLC is one of the simplest techniques routinely used in pharmaceutical quality control laboratories because of its sensitivity, speed, selectivity, and ease of performance. Several HPLC procedures [24-31] employing different columns and mobile phases have been reported for its assay in dosage forms when present alone [24-30] or in combination with bezafibrate [31]. Tagliari et.al [24] used methanol and water in the ratio of 95: 5 is used as mobile phase at flow rate of 1.2 mL min^{-1} on a C-18 column. In a method reported by Florea et.al [25] buffer consisting of 1-heptanesulfonate sodium salt was used as ion pair and acetonitrile in the ratio 60:40 (v/v) as mobile phase. C-18 column was used as stationary phase and 220nm is wavelength of detection. Gopal et.al [26] reported RP-HPLC method for the estimation of TFH in dosage forms on C-18 column by using buffer and acetonitrile (65:35 v/v) as mobile phase at aflow rate of 1.8 mL min-1. Rani et.al [27]

reported another method for estimation of TFH in tablets by using ODS column as stationary phase, acetonitrile and phosphate buffer (40:60 v/v) as mobile phase. Simultaneous determination of TFH and its four impurities by HPLC has been reported by Matysova et.al [28] by using tetrahydrofuran, acetonitrile and phaosphate buffer as mobile phase. Another method for estimation of TFH in the presence of its photo degradation products was reported by Abdel-Moety et.al [29] by using methanol and water (80:20 v/v) as mobile phase on a C-18 column. Cardoso S G and Schapoval E E reported hplc method for the estimation of TFH in tablets and creams [30]. Simultaneous determination of TFH and bezafibrate is reported by Ramesh raju and Bujji babu by using C-18 column as stationary phase and methanol, acetonitrile, phosphate buffer as mobile phase [31].

Stability indicating assays are increasingly being applied to many pharmaceutically important compounds [32]. But, none of the methods available for TFH [9-11,24-27,29-31] is stability indicating. Many reported HPLC methods require expensive solvents like acetonitrile [25-28,31], corrosive solvents like tetrahydrofuran [28], and ion pairing reagents [25]. The objective of the present study was to develop a simple, rapid, selective, precise, accurate, and stability indicating HPLC method for the determination of TFH in pharmaceuticals. The assay was performed on a Zorbax Eclips XDB C-18 column and with a mobile phase consisting of buffer (1000 mL water, 2 mL triethylamine, pH 3.4 adjusted with trifluoroacetic acid.), isopropyl alcohol and methanol (40:12:48, v/v/v), at a flow rate of 1 mL min^{-1} and UV detection at 222nm. The proposed method was successfully applied to the determination of TFH in its tablets by using an inexpensive mobile phase.

MATERIALS AND METHODS

Chemicals and reagents

Pharmaceutical grade TFH was received from Dr. Reddy's laboratories limited, Hyderabad, India, as gift sample. Two brands of tablets, Zimig-250 (from Glaxo Smith Kline Pharmaceuticals Limited, India) and Terbiforce-250 (from Lifestar Pharma PVT. Ltd.) used in the investigation were purchased from commercial sources in the local market. All the reagents and chemicals used were of analytical reagent grade. Triethyl amine (99%), and trifluoro acetic acid (99%) were obtained from Merck, Mumbai, India. HPLC grade iso propyl alcohol and methanol were obtained from Merck, Mumbai, India.

Buffer was prepared by mixing 2.0 mL of triethylamine with 1000 mL of water and the pH was brought to 3.4 by adding dilute trifluoro acetic acid using a pH meter. All solutions were

prepared in bi-distilled water. Then mobile phase was prepared by mixing buffer, isopropyl alcohol and methanol in the ratio 40:12:48 v/v/v.

Instrumentation and chromatographic conditions

Chromatographic analysis was carried out using Alliance Waters HPLC system equipped with Alliances 2695 series low pressure quaternary pump with a programmable Waters 2998 photodiode array detector and auto sampler. Data were collected and processed using Waters Empower 2.0 software. The separation was performed using an Zorbax Eclips XDB C-18 column (3.5 µm, 4.6 × 150 mm i.d.). The mobile phase was composed of buffer (1000 mL water, 2 mL triethylamine, pH 3.4 adjusted with trifluoroacetic acid.), isopropyl alcohol and methanol (40:12:48, v/v/v). The flow rate was maintained at 1.0 mL min^{-1}. The column effluent was monitored on UV detector set at 222 nm.

GENERAL PROCEDURES

Procedure for preparation of solutions

Preparation of standard solution.

A TFH standard stock solution (200 µg mL^{-1}) was prepared by dissolving 20 mg of the pure drug in 100 mL of methanol. Working solution of 40 µg mL^{-1} were prepared by diluting 2.0 mL of the standard stock solution to 10 mL with the methanol in a calibrated flask.

Preparation of sample solution.

A TFH sample stock solution (200 µg mL^{-1}) was prepared by dissolving 20 mg of the pure drug in 100 mL of methanol. Working solution of 40 µg mL^{-1} were prepared by diluting 2.0 mL of the sample stock solution to 10 mL with the methanol in a calibrated flask.

Procedure for preparation of calibration curve

Working solutions equivalent to 1.0 - 80.0 µg mL^{-1} TFH were prepared by diluting appropriate aliquots of the stock solution. Aliquots of 10 µL were injected (triplicate) and eluted with the mobile phase under the reported chromatographic conditions. The average peak area versus the concentration of TFH in µg mL^{-1} was plotted (Figure-1). Alternatively, corresponding regression equation was derived using the mean peak area-concentration data and the concentration of the unknown was computed from the regression equation.

Procedure for tablets

Twenty tablets were accurately weighed, finely pulverized and mixed using a mortar and pestle. An amount of tablet powder equivalent to 20 mg of TFH was weighed and transferred into a 100 mL volumetric flask, 50 mL of methanol was added and was sonicated for 20 min in an ultrasonic bath to complete dissolution of the TFH, the content was then diluted to the mark with

the methanol, mixed well and filtered using a 0.22µm nylon membrane filter paper. Aliquots of this solution were successively diluted with the methanol and then subjected to analysis as per the general procedure described for the calibration curve.

Procedure for placebo blank analysis

A placebo blank of the composition: Lactose (10mg) talc (50 mg), starch (50 mg), acacia (50mg), methyl cellulose (20 mg), sodium citrate (20 mg), magnesium stearate (20 mg) and sodium alginate (10 mg) was made and its solution was prepared as described under 'Procedure for tablets', by taking about 40 mg and then analyzed using the procedure described earlier.

Procedure for the determination of TFH in synthetic mixture

To ~ 20 mg of the placebo blank of the composition described above, 20 mg of TFH was added and homogenized, transferred to a 100 mL calibrated flask and the solution was prepared as described under "Procedure for tablets", and then subjected to analysis by the procedure described above. This analysis was performed to study the interference by excipients such as lactose, talc, starch, acacia, methyl cellulose, sodium citrate, magnesium stearate and sodium alginate.

Preparation of acid, base and water hydrolysis induced-degradation of sample

For acid, alkaline and water hydrolysis degradation studies to 20 mg of pure TFH drug, 2.5 mL of 2M HCl or 2M NaOH or water were added separately in three 100 mL calibrated flasks. The flasks were kept on a water bath set at 80^{o}C for 3.0 hrs, then cooled to room temperature. Then the solutions were neutralized with equal volume of 2M NaOH or 2M HCl. The content of each flask was made up to the mark with methanol. Further diluted these degraded samples to get 40 µg mL^{-1} with methanol. Aliquots of 10 µL of each degraded sample were injected (duplicate) and eluted with the mobile phase under the reported chromatographic conditions.

Preparation of hydrogen peroxide induced-degradation of sample

To 20 mg of pure TFH drug 2.5 mL of 3% hydrogen peroxide was added in 100 mL calibrated flask and kept on a water bath set at 80^{0}C for 3.0 hrs. The flask was cooled to room temperature and made up to the mark with methanol. Further this degraded sample is degraded to get 40 µg mL^{-1} with methanol. Injected 10 µL of degraded sample in duplicate and eluted with the mobile phase under the reported chromatographic conditions.

Preparation of dry heat degradation and photo-degradation samples.

The powdered drug was stored in the oven at 105^{0}C for 48 hrs to study dry heat degradation, and exposed to 200 watt hr. m^{-2} UV-radiation and 1.2 million lux hr. of visible radiation for study of photo degradation. Then, solutions containing 40 µg mL^{-1} TFH in methanol were prepared

separately and 10 µL of degraded sample was injected in duplicate and eluted with the mobile phase under the reported chromatographic conditions.

RESULTS AND DISCUSSION

Method development and optimization

The proposed method permits the quantitation of TFH in the pure drug and commercial tablets in the presence of its degradation products. In order to obtain good linearity, sensitivity and selectivity, the method was optimized and validated in accordance with the current ICH guidelines [33]. A well defined symmetrical peak and good results were obtained upon measuring the response of eluent under the optimized conditions after thorough experimental trials that could be summarized as follows:

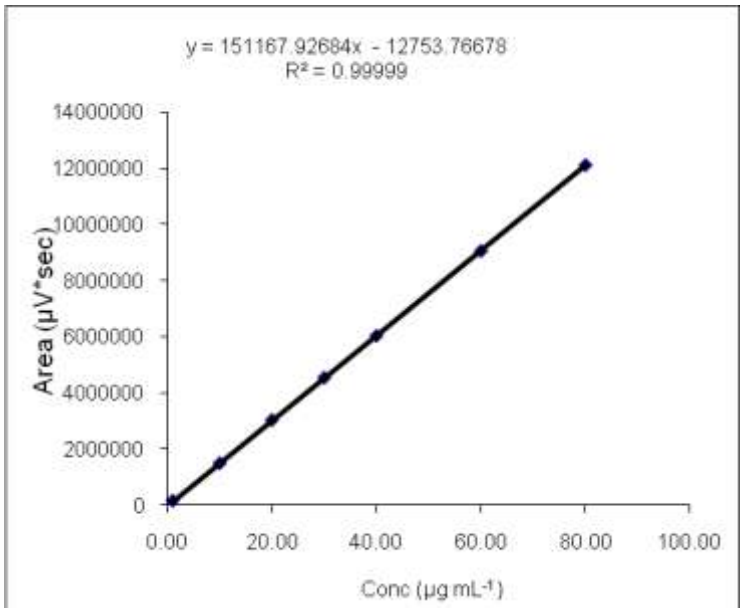

Figure 1: Calibration graph

Choice of column

Three different columns were used for performance investigations, including hypersil BDS C_8 (250 mm x 4.0 mm i.d, 5.0 µm particle size) thermo column, Intersil C_{18} (150mm × 3.9 mm i.d., 5 µm,) column and Zorbax Eclips XDB C-18 (150 mm x 4.6 mm i.d., 3.5 µm particle size) column. The studies revealed that the Zorbax Eclips XDB C-18 column was more suitable since it gave better sensitivity.

Choice of wavelength

Spectroscopic analysis of the compound with PDA detector showed that TFH has maximum UV absorbance at 222 nm (Figure.-2). Therefore, the chromatographic detection was performed at 222 nm.

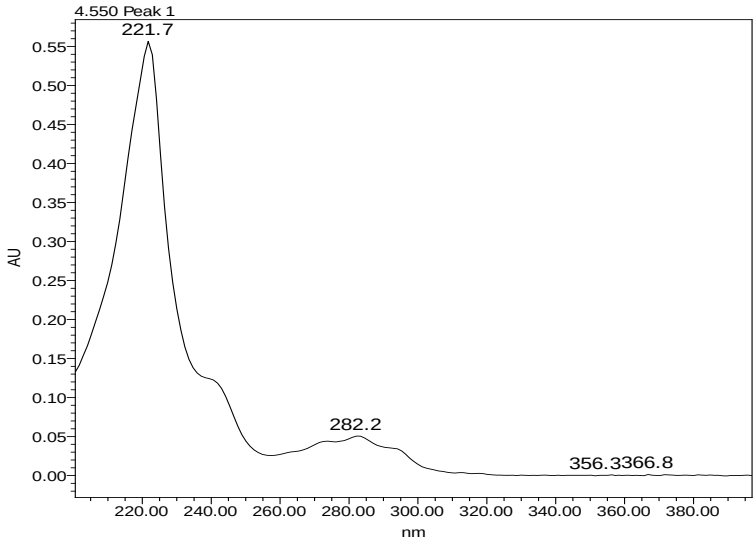

Figure 2 (Absorption spectrum of the eluted species)

Mobile phase composition

Chromatographic conditions were optimized by changing the mobile phase composition and buffers used in the mobile phase. Different experiments were carried out to optimize the mobile phase. Several modifications in the mobile phase composition were performed in order to study the possibilities of changing the selectivity of the chromatographic system. These modifications included the change of the type and ratio of the organic modifier, the pH, the strength of the buffer, and the flow rate. Precise and accurate results with maximum number of theoretical plates and good peak shape were obtained when the mobile phase consisting of buffer (1000 mL water, 2 mL triethylamine, pH 3.4 adjusted with trifluoroacetic acid.), isopropyl alcohol and methanol (40:12:48, v/v/v) was used.

Type of organic modifier

Acetonitrile, methanol and isopropyl alcohol were used separately as organic modifiers during development but they did not give a good peak. Methanol and isopropyl alcohol combination was the organic modifier of choice which gave a nice, elegant and highly sensitive peak.

Ratio of organic modifier

The effect of ratio of organic modifier on the selectivity and retention time of the test solute was investigated using different mobile phase compositions. At buffer, isopropyl alcohol and methanol (40:12:48, v/v/v) ratio well defined peak and the highest number of theoretical plates were observed.

Effect of pH and ionic strength of buffer

The effect of pH of the mobile phase on the selectivity and retention time of the test solute was investigated using mobile phases of pH ranging from 2.5 – 8.0. The results revealed that pH 3.4

was most appropriate giving well defined peak and the highest number of theoretical plates. At lower and higher pH, non-symmetrical peak and smaller number of theoretical plates were observed. Therefore pH 3.4 was fixed as optimum. The same trend was observed with respect to the ionic strength of the buffer and not much change was observed with increase in the ionic strength of the buffer, and hence, ~ 2.0 mL triethyl amine buffer was used as working buffer throughout the investigation.

The effect of flow rate

The effect of flow rate on the symmetry, sensitivity and retention time of the peak was studied and a flow rate of 1 mL min-1 was optimal for better symmetry and reasonable retention time.

Stability study

In the present work, high performance liquid chromatographic method for the determination of TFH in bulk drug and in tablets have been described. Methanol used for sample preparation (Figure 3) and TFH standard solution (Figure 4) injected and eluted with the mobile phase under the reported chromatographic system.

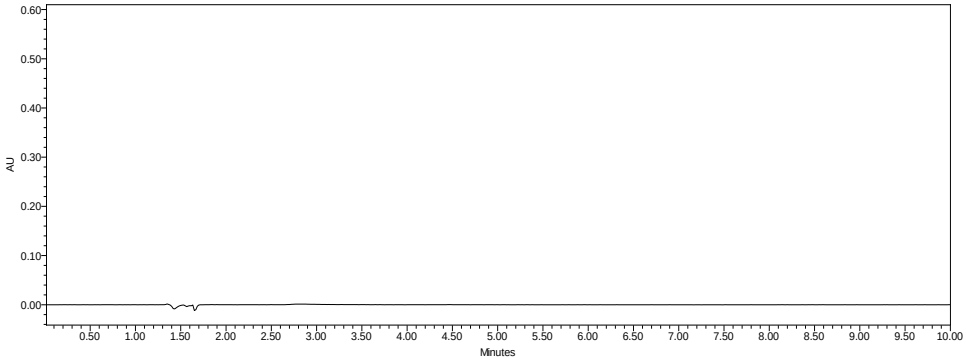

Figure 3: Blank Chromatogram.

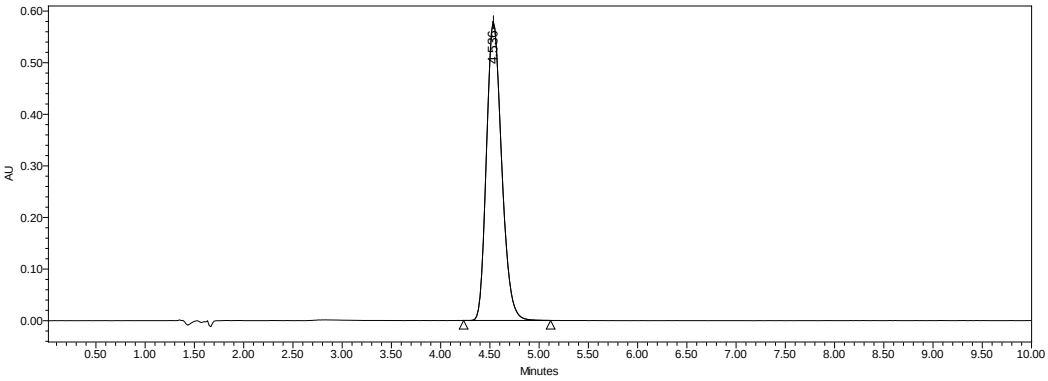

Figure 4: Standard Chromatogram (40 µg mL^{-1}).

TFH samples treated with acid, base, water, hydrogen peroxide, dry heat and UV-Visible radiation were injected (duplicate) and eluted with the mobile phase under the reported

chromatographic conditions. The degradation study was based on the comparison of the TFH peak area of "stressed TFH samples" with that of the "standard TFH solution".

On comparison of TFH stressed sample chromatograms with that of the un-stressed it can concluded that TFH is quite stable under acid, base, water, dry heat and photo stress conditions (Figure 5-9), and unstable under oxidative stress condition (Figure 10).

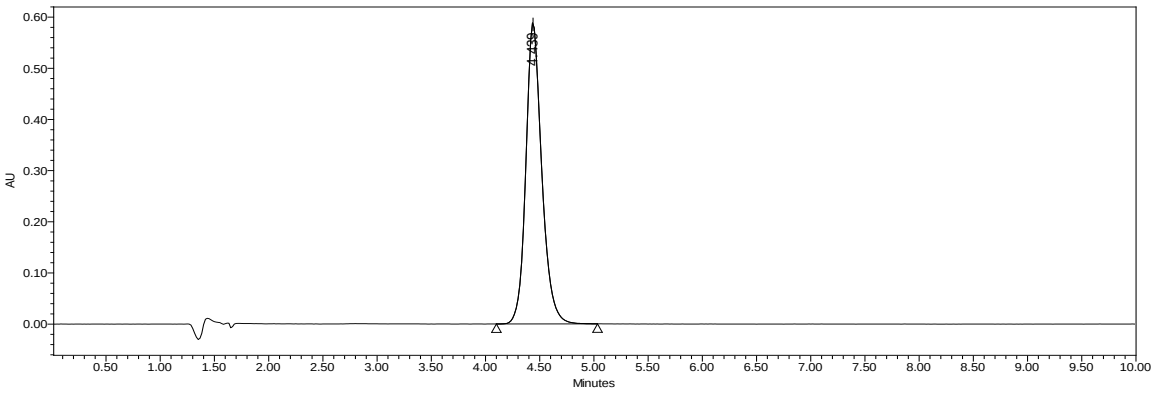

Figure 5: 40 µg mL^{-1} Sample (Acid Deg-2M HCL/80°C/3Hrs)

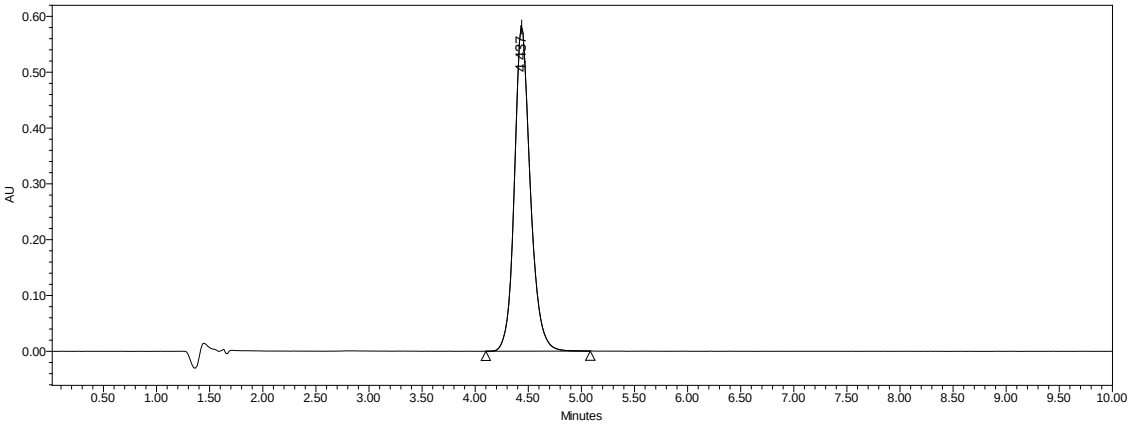

Figure 6: 40 µg mL^{-1} Sample (Base Deg-2M NaOH/80°C/3Hrs)

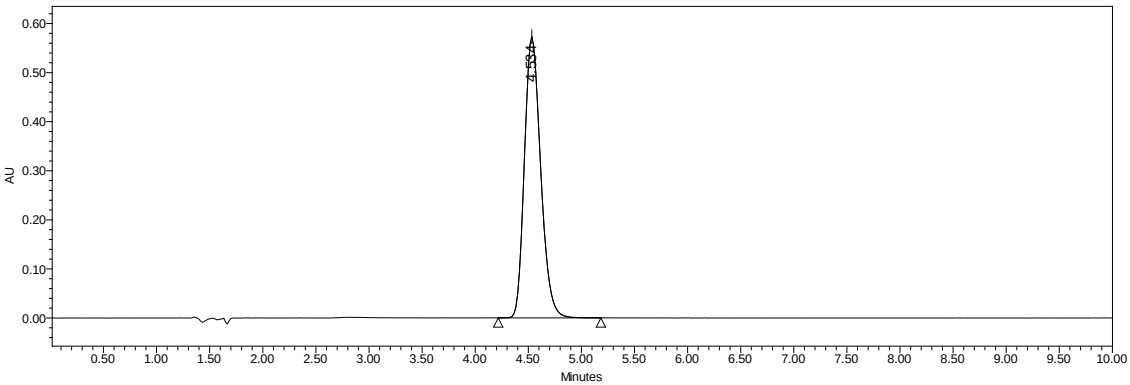

Figure 7: 40 µg mL^{-1} Sample (Hydrolysis- Water/80°C/3Hrs)

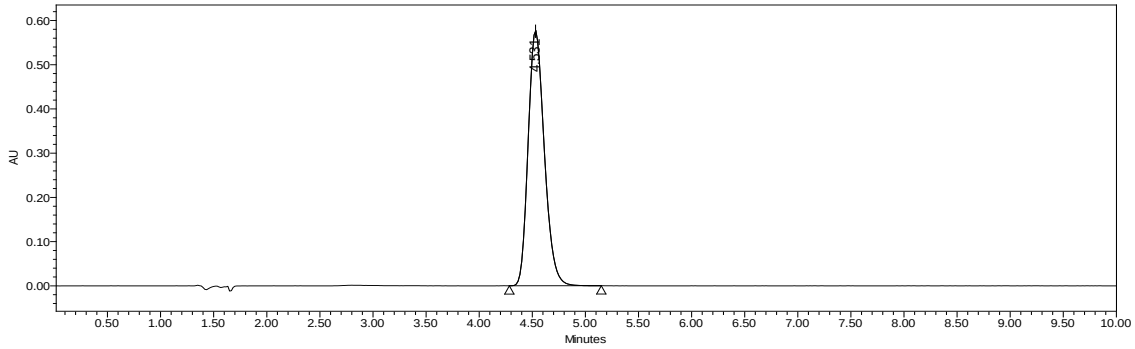

Figure 8: 40 µg mL^{-1} Sample (Thermal Degradation-105°C / 48Hrs)

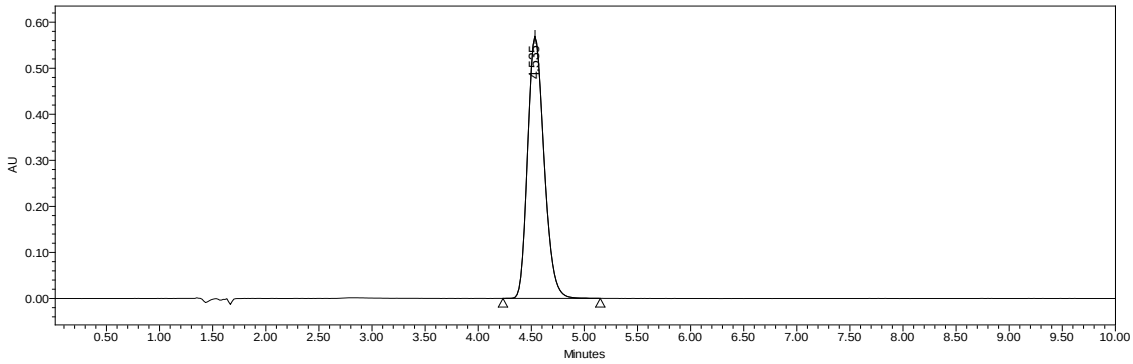

Figure 9: 40 µg mL^{-1} Sample (Photolytic Degradation, UV-200 Whr, Visible-1.2M lux hr)

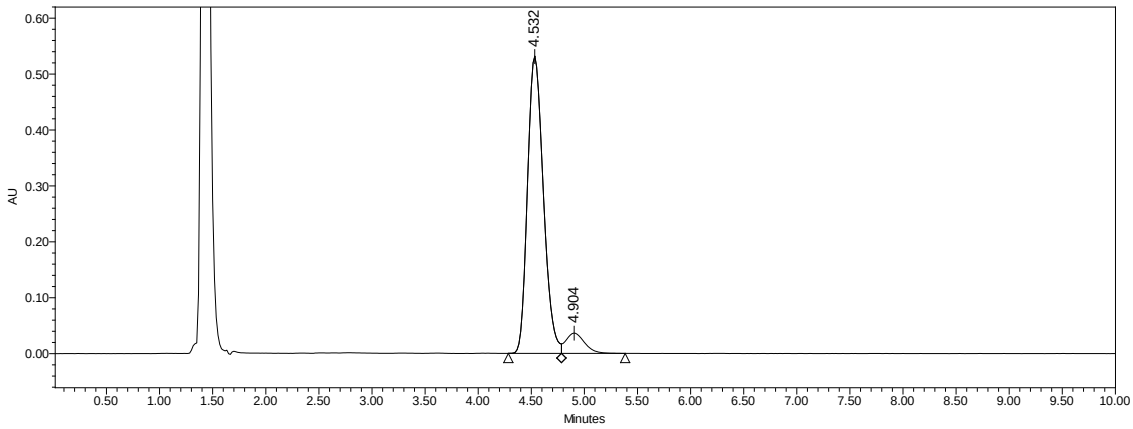

Figure 10: 40 µg mL^{-1} Sample (Peroxide Deg-5%H_2O_2/80°C/3Hrs)

VALIDATION OF THE METHOD

Linearity

Working standard solution of TFH (200 µg mL^{-1}) was appropriately diluted with the methanol to obtain solutions in the concentration range 1 - 80 µg mL^{-1} TFH. Ten microlitre of each solution was injected in triplicate onto the column under the operating chromatographic conditions described above. The least squares method was used to calculate the slope, intercept and the correlation coefficient (r) of the regression line. The relation between mean peak area Y (n=3)

and concentration, X expressed by the equation Y = 151167.92684 X + 12753.76678 (r^2 = 0.9999), was linear. Related statistical data are presented in Table 1.

Table 1: Sensitivity and regression parameters

Parameter	Value
Linearity range, µg mL^{-1}	1-80
Regression (Y^* = a ± bX)	
Slope (b)	151167.9268
Intercept (a)	-12753.76678
Standard deviation of intercept (S_a)	134765.37
Standard deviation of Slope (S_b)	2895.224579
Correlation co-efficient (r)	0.9999
Limit of detection (LOD, µg mL^{-1})	0.3
Limit of quantification (LOQ, µg mL^{-1})	1

*Y = a+bX, where Y is the area and X concentration in µg mL^{-1}.

Limits of quantification (LOQ) and detection (LOD)

The limit of quantification (LOQ) was determined by establishing the lowest concentration that can be measured according to ICH recommendations [20], below which the calibration graph is non linear and was found to be 1µg mL^{-1}. The limit of detection (LOD) was determined by establishing the minimum level at which the analyte can be reliably detected and it was found to be 0.3 µg mL^{-1}.

Selectivity

A systematic study was performed to determine the effect of matrix by analyzing the placebo blank and synthetic mixture containing TFH. Method selectivity was checked by comparing the chromatograms obtained for placebo blank (Figure 11), pure TFH solution (Figure 2), synthetic mixture and tablet solution (Figure 12,13).

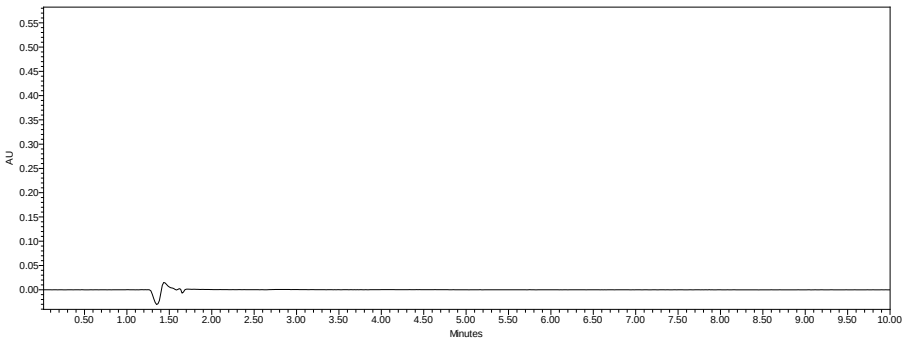

Figure 11: Placebo Blank.

An examination of the chromatograms of the above solutions revealed the absence of peaks due to additives present in tablet preparations. The peak area value resulting from 40 µg mL^{-1} TFH in synthetic mixture solution had nearly the same as that obtained for pure TFH solutions of

identical concentration. This unequivocally demonstrated the non-interference of the inactive ingredients in the assay of TFH. Further, the slopes of the calibration plots prepared from the synthetic mixture solutions were about the same as those prepared from pure drug solutions.

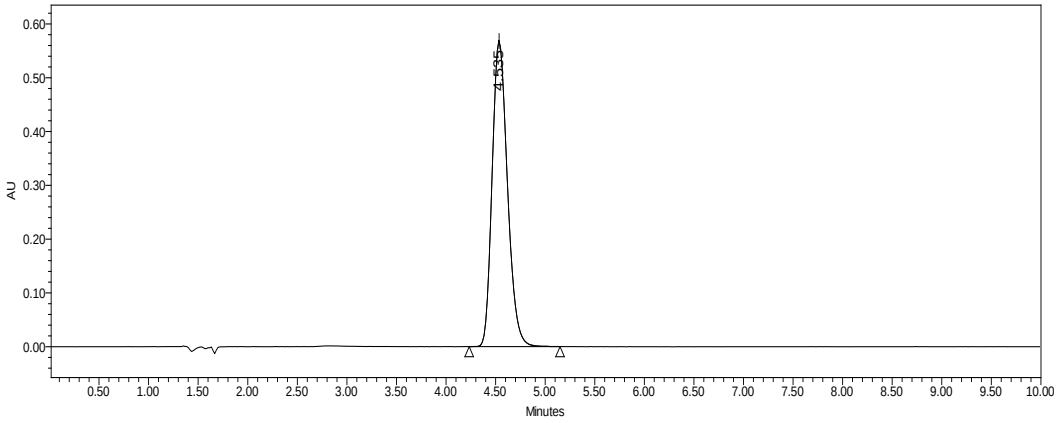

Figure 12: Tablet Chromatogram.

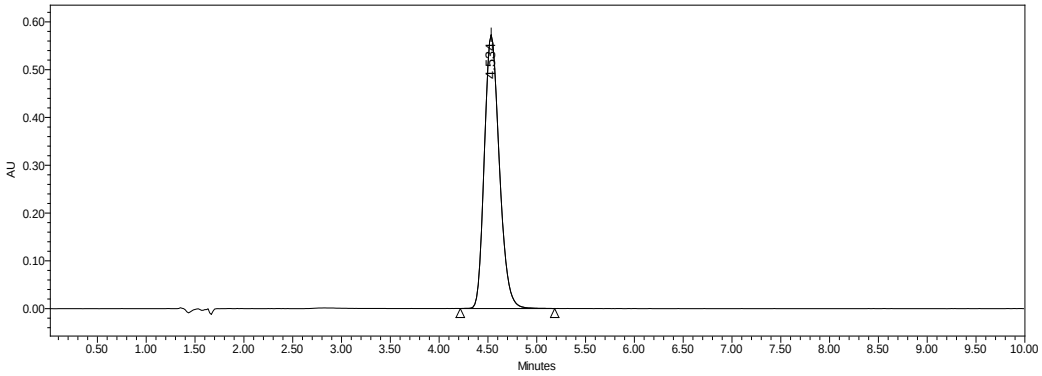

Figure 13: Synthetic mixture Chromatogram.

Precision and Accuracy

Method precision was evaluated from the results of seven independent determinations of TFH at three different concentrations, 20.0, 40.0 and 60.0 µg mL^{-1} TFH on the same day and on five consecutive days. The inter-day and intra-day relative standard deviation (RSD) values for peak area and retention time for the selected concentrations of TFH were less than 1%. The method accuracy, expressed as relative error (%) was determined by calculating the percent deviation found between concentrations of TFH injected and concentrations found from the peak area.

This study was performed by taking the same three concentrations of TFH used for precision estimation. The intra-day and inter-day accuracy (expressed as %RE) was less than 1% and the values are compiled in Table 2.

Table 2: Intra-day and Inter-day Accuracy and Precision.

TFH Injected, µg mL^{-1}	Intra-day accuracy and precision				Inter-day accuracy and precision			
	TFH found[a]	%RE	%RSD[b]	%RSD[c]	TFH found[a]	%RE	%RSD[b]	%RSD[c]
20	20.13	0.65	0.23	0.05	20.07	0.35	0.19	0.25
40	40.19	0.47	0.83	0.11	40.06	0.15	0.71	0.30
60	59.92	-0.13	0.03	0.06	60.07	0.12	0.09	0.27

a-Mean value of seven determinations, b-Based on peak area, c-Based on retention time

Robustness and Ruggedness

To determine the robustness of the method small deliberate changes in the chromatographic conditions like column temperature, mobile phase buffer pH, flow rate, and detection wavelength were made, and the results were compared with those of the optimized chromatographic conditions. In each case the %RSD values were calculated for the obtained peak area and retention time. The results of this study expressed as %RSD are summarized in Table 3.

Table 3: Robustness and ruggedness study (TFH concentration, 40 µg mL-1, n = 3)

Condition	Modification	Mean peak area ±S D	%RSD	Mean Rt ± SD	%RSD
Optimized Condition	-----	6156092 ± 18872	0.31	4.539 ± 0.005	0.11
Temperature (ºC)	29	6467947 ± 43613	0.67	4.552 ± 0.004	0.08
	31	6562809 ± 42958	0.65	4.470 ± 0.003	0.07
Mobile phase Buffer pH	pH-3.3	6520710 ± 48261	0.74	4.728 ± 0.006	0.12
	pH-3.5	6035841 ± 42589	0.71	4.310 ± 0.005	0.12
Flow rate (mL min^{-1})	0.9	6760583 ± 28487	0.42	4.987 ± 0.004	0.09
	1.1	6599215 ± 22926	0.41	4.117 ± 0.005	0.12
Wavelength (nm)	221	6532646 ± 59285	0.91	4.504 ± 0.005	0.12
	223	6670806 ± 56723	0.85	4.498 ± 0.006	0.13
Different Analyst	Analyst - 1	6257102 ± 33685	0.54	4.506 ± 0.004	0.09
	Analyst - 2	6353221 ± 41436	0.65	4.611 ± 0.005	0.11
	Analyst - 3	6293681 ± 42865	0.68	4.583 ± 0.005	0.11
Different Column	Column-1	6361581 ± 51351	0.81	4.443 ± 0.004	0.09
	Column-2	6358415 ± 54662	0.86	4.613 ± 0.006	0.13
	Column-3	6158453 ± 48351	0.79	4.588 ± 0.004	0.09
Different Day	Day-1	6284165 ± 44387	0.71	4.538 ± 0.004	0.09
	Day-2	6158546 ± 51384	0.83	4.558 ± 0.005	0.11
	Day-3	6215864 ± 49358	0.79	4.511 ± 0.004	0.09

At the deliberate varied chromatographic conditions (flow rate, temperature, and mobile phase buffer pH and detector wavelength), the analyte peak area and retention time %RSD remained near to the actual values. The RSD values ranged from 0.31 to 0.91% resumes the robustness of the proposed method. To study the ruggedness of the method three different columns of same dimensions were used for the analyses. The studies were performed on the same day and on three different days by three different analysts for three different concentrations of TFH (triplicate

injections). In each case the %RSD values were calculated for the obtained peak area and retention time. The results of this study expressed as %RSD are summarized in Table 3. The analyte peak area and retention time %RSD remained closer to the values under optimized condition.

Application to tablets.

The developed and validated method was applied to the assay of TFH commercial tablets. The results shown in Table 4 are in good agreement with the label claim and with those obtained with the reference method [7]. The reference method involved titration with standard NaOH in hydro-alcoholic medium, the end point being located potentiometrically. The results showed that the Students' t- and F-values at 95% confidence level did not exceed the tabulated values, which confirmed that there is a good agreement between the results obtained by the proposed methods and the reference method with respect to accuracy and precision. Figure 4 shows a chromatogram indicating good peak due to TFH from the tablet. Therefore, the proposed method can be used for the quality control of the tablets.

Recovery study

To further assess the accuracy and reliability of the method, recovery studies *via* standard addition method was performed. To the pre-analyzed tablet powder, pure TFH was added at three levels and the total was found by the proposed method. Each test was triplicated. When the test was performed on two different brands of tablets, the percent recovery of pure TFH was in the range of 99.4 – 99.8 with standard deviation values of 0.07 – 0.22. The results indicated that the method is very accurate and that common excipients found in tablet preparations did not interfere. The results are complied in Table 5.

Table 4: Results of determination of TFH in tablets and statistical comparison with the reference method.

Tablet brand name	Nominal amount, mg	Found* (Percent of label claim ± SD) Reference method	Proposed method
Zimig	250	98.6 ± 1.13	99.38 ± 0.50 t = 1.42 F = 5.12
TERBIFORCE	250	98.8 ± 1.10	99.98 ± 0.54 t = 2.14 F = 4.15

*Mean value of five determinations

Table 5: Results of recovery study by standard addition method

Tablet	TFH in tablet,	Pure TFH	Total	Pure	TFH

	µg mL^{-1}	**added, µg mL^{-1}**	**found, µg mL^{-1}**	**recovered*, Percent ± SD**
	19.88	10	29.96	100.3 ± 0.221
Zimig	19.88	20	39.84	99.9 ± 0.090
	19.88	40	59.77	99.8 ± 0.027
	20	10	29.91	99.7 ± 0.070
TERBIFORCE	20	20	39.88	99.7 ± 0.197
	20	40	59.85	99.8 ± 0.132

*Mean value of three determinations

CONCLUSION

The isocratic RP-HPLC method developed for quantitative analysis of terbinafine hydrochloride in pharmaceutical dosage forms is precise, accurate, robust and specific. Satisfactory results were obtained from validation of the method. The retention time obtained (4.5 min) enables rapid determination of the drug which is important in routine analysis. The method exhibited an excellent performance in terms of sensitivity and speed. The method is stability indicating and can be used for routine analysis of production samples and can be used for the assay of terbinafine either in pure drug or pharmaceutical formulations. Degradation study of the method reveals that the product is unstable in peroxide medium. The proposed method is superior over many other reported methods in terms of stability indicating nature which is most important requirement for an assay method, inexpensive solvents were used in mobile phase with less solvent composition, lower flow rate and with less run time. No corrosive solvents or ion pairing reagents were used which lead to longer column equilibration time and reduces the column life.

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Conflict of interest.

We do not have any conflict of interest and any financial assistance with the commercial identities mentioned in the article.

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