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Development and Validation of Analytical Method for Estimation of Terbinafine by Using UV Spectroscopy

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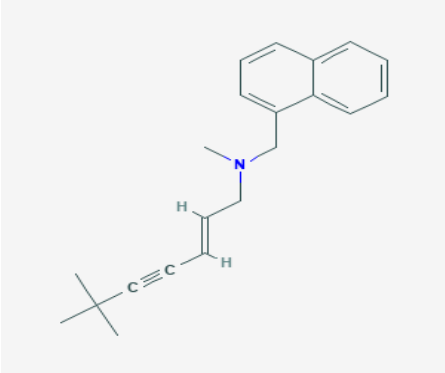
ABSTRACT

Background

The main objective was to develop and validate the UV-spectrophotometric method for the estimation of terbinafine hydrochloride in pure pharmaceutical formulations as per ICH guidelines. **Materials and Methods:** A simple, rapid, accurate, and economical UV-spectrophotometric method has been developed for the estimation of terbinafine hydrochloride from bulk and pharmaceutical formulation. **Results:** The λ_{max} of terbinafine hydrochloride in methanol was found to be 224 nm. The drug follows linearity in the concentration range 0.4–2.8 µg/ml with a correlation coefficient value of 0.998. The proposed method was applied to pharmaceutical formulation and % amount of drug estimated was 99.58% and was found to be in good agreement with the label claim. The accuracy of the method was checked by a recovery experiment performed at three different levels, i.e., 80%, 100%, and 120%. The % recovery was found to be in the range of 99.58– 102.78%. The low values of % RSD are indicative of the accuracy and reproducibility of the method. The precision of the method was studied as an intraday; interday variations, and repeatability. The % RSD value < 2 indicates that the method is precise. The ruggedness of the proposed method was studied with the help of two analysts. **Conclusion:** The above method was a rapid tool for routine analysis of terbinafine hydrochloride in the bulk and the pharmaceutical dosage form.

INTRODUCTION:

Terbinafine hydrochloride(*E*)-*N*-(6,6-dimethyl-2-hepten-4-ynyl)-*N*-methyl-1-naphthalene methanamine hydrochloride.



It's a new potent antifungal agent of the allylamine class that selectively inhibits fungal *squalene epoxidase*. The drug is indicated for both oral and topical treatment of mycoses. Terbinafine hydrochloride is not yet official in any pharmacopeia, where, only a few analytical methods have been reported for its determination in pharmaceutical formulations and biological fluids. Such methods include HPLC, colorimetry, electrochemistry, and solvent meting method. Among the various methods available for the determination of drugs, spectrophotometry continues to be very popular, because of its simplicity, specificity, and low cost. This study presents a new spectrophotometric method for the determination of terbinafine hydrochloride phosphate in bulk and pharmaceutical formulations. Accordingly, the objective of this study was to develop and validate the UV-spectrophotometric method for the estimation of terbinafine hydrochloride in bulk and pharmaceutical formulations as per ICH guidelines.

MATERIAL AND METHODS:

Materials

Terbinafine hydrochloride was a gift sample from Sun pharmaceutical Baddi. All chemicals and reagents used were of analytical grade.

Preparation of standard stock solution

10 mg of drug was weighed accurately and 10 ml methanol was transferred into a volumetric flask and sonicated for 5 min. Then a standard stock solution of Terbinafine (10µg/ml) was prepared with 100ml methanol.

Selection of wavelength for analysis of terbinafine hydrochloride

The standard stock solution was further diluted with methanol, to obtain various dilutions from 0.4-2.8µg/ml. The absorbance of these solutions was recorded at 224nm against methanol as blank using a UV-visible spectrophotometer and a standard curve was plotted against concentration. From the calibration curve intercept, slope, straight-line equation, and correlation coefficient were obtained.

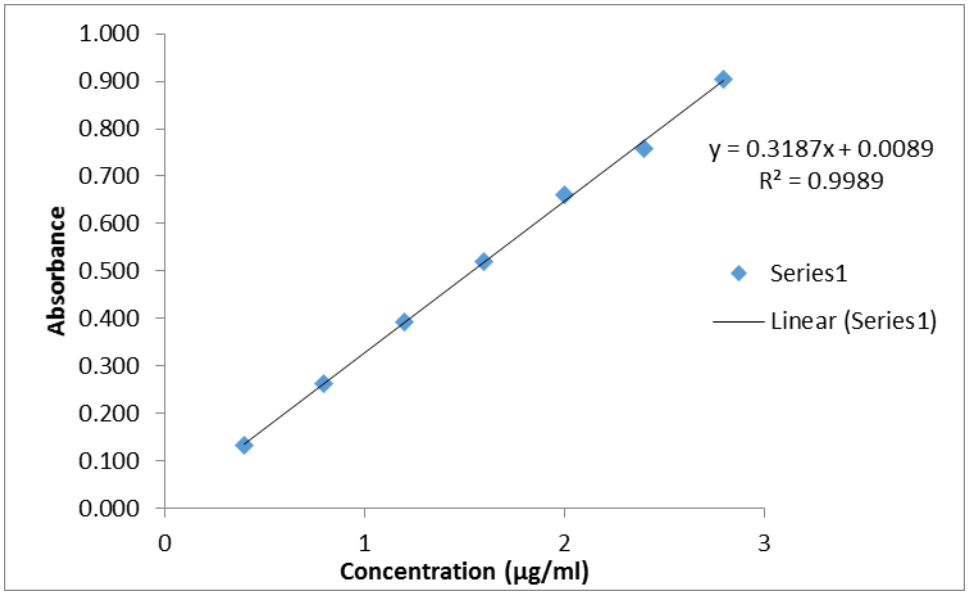


Figure No 1: Graph of standard calibration curve of Terbinafine in methanol (λ_{max}= 224nm)

1. Validation of the method

The method was validated in terms of linearity, accuracy, precision, and ruggedness.

a) Linearity

A calibration curve was plotted over a concentration range of 0.4 to 2.8µg/ml for Terbinafine at 224nm. Accurately measured working stock solution of Terbinafine at 224nm (0.4, 0.8, 1.2, 1.6, 2.0, 2.4, 2.8ml) were transferred to a separate series of 10ml volumetric flask and diluted up to the mark with methanol. The absorbance of all solutions was taken at their respective wavelength. The Linearity was constructed by plotting concentration against absorbance were each reading.

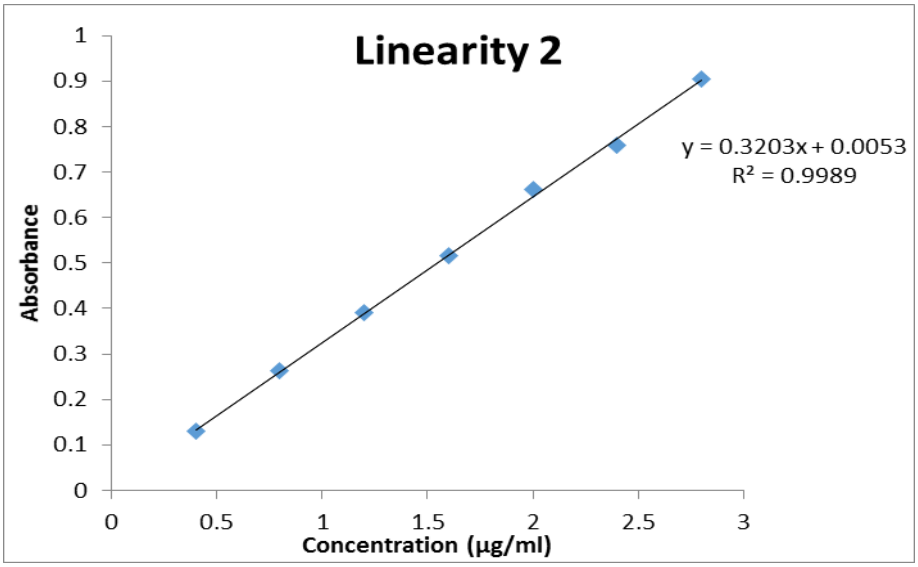


Figure No. 2: Linearity graph of Terbinafine

b) Precision

The term precision is defined by the ISO International Vocabulary of Basic and General Terms in Metrology (ISO-VIM) and ICH as the closeness of agreement between quantity values obtained by replicate measurements of a quantity under specified conditions. Assessing the precision implies expressing numerically the random error or the degree of dispersion of a set of individual measurements using the standard deviation, the variance, or the coefficient of variation.

c) Repeatability

It is the concordance of a series of measurements of the same quantity when the experiments are conducted under the same conditions (analyst, apparatus, instrument, and day) in rapid succession. For this experiment, a standard solution of Terbinafine (1.6 μg/ml) was prepared and analyzed six times as per the proposed method.

d) Accuracy

Accuracy was determined using recovery experiments, by the determination of % mean recovery of the sample at three different levels (80-120%). At each level, three determinations were performed. The accepted limits of recovery are 90% - 120% and all observed data are within the required range which indicates good recovery values and hence the accuracy of the method developed.

e) Ruggedness

Ruggedness was determined by carrying out analysis by two different analysts and the respective percentage recovery was noted and the results were indicated as % RSD.

f) Sensitivity

The detection limit of an individual analytical procedure is the lowest amount of analyte in the sample which can be detected but not necessarily quantitated as an exact value. The Quantitation limit of an individual analytical procedure is the lowest amount of analyte in the sample which can be quantitatively determined with suitable precision and accuracy. The LOD and LOQ of the proposed method were determined by using a calibration curve:

$$\mathrm{LOD} = \frac{3.3\sigma}{S}, \qquad \mathrm{LOQ} = \frac{10\sigma}{S},$$

Where σ is the standard deviation of the response (Y-intercept) and S is the slope of the calibration curve.

2. Validation of UV Spectroscopy Method

a) Linearity

A calibration curve was plotted over a concentration range of 0.4 to 2.8µg/ml for Terbinafine at 224nm. Accurately measured working stock solution of Terbinafine at 224nm (0.4, 0.8, 1.2, 1.6, 2.0, 2.4, 2.8ml) were transferred to a separate series of 10ml volumetric flask and diluted up to the mark with methanol. The absorbance of all solutions was taken at their respective wavelength. The Linearity was constructed by plotting concentration against absorbance were each reading.

Table no 1: Linearity of Terbinafine

Sr. No.	Concentration µg/ml	Linearity 1	Linearity 2	Linearity 3
		Absorbance	Absorbance	Absorbance
1	0.4	0.132	0.13	0.136
2	0.8	0.263	0.262	0.265
3	1.2	0.394	0.391	0.391
4	1.6	0.523	0.517	0.524
5	2	0.662	0.661	0.661
6	2.4	0.757	0.758	0.759
7	2.8	0.9	0.905	0.905

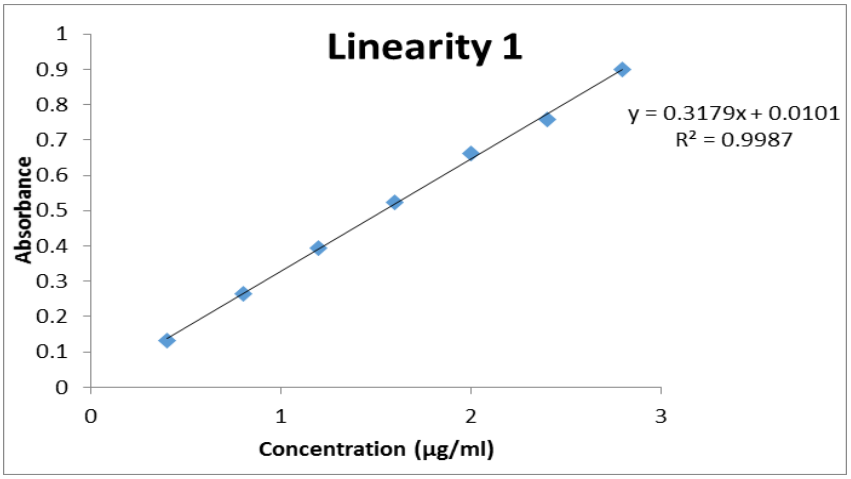


Figure No. 3: Linearity 1 graph of Terbinafine

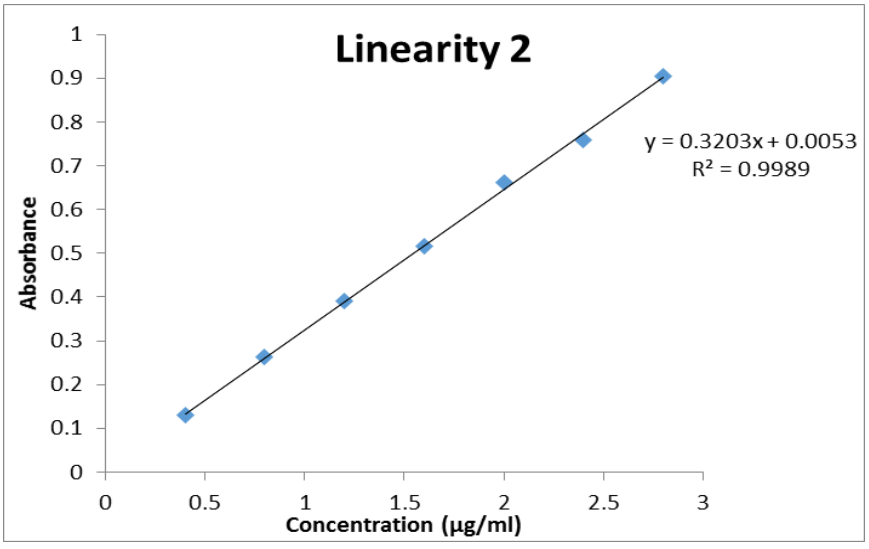


Figure No. 4: Linearity 2 graph of Terbinafine

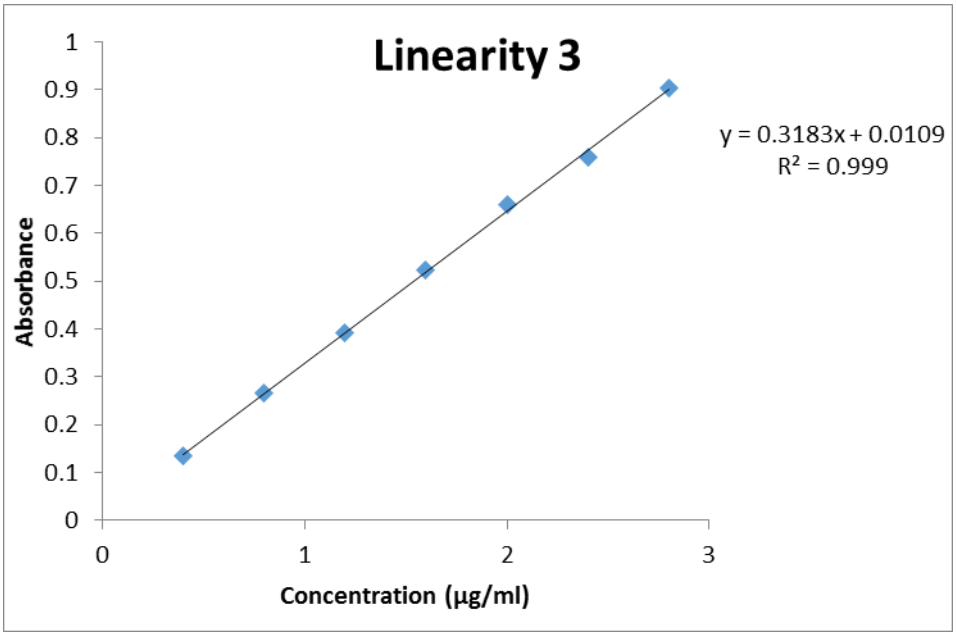


Figure No. 5: Linearity 3 graph of Terbinafine

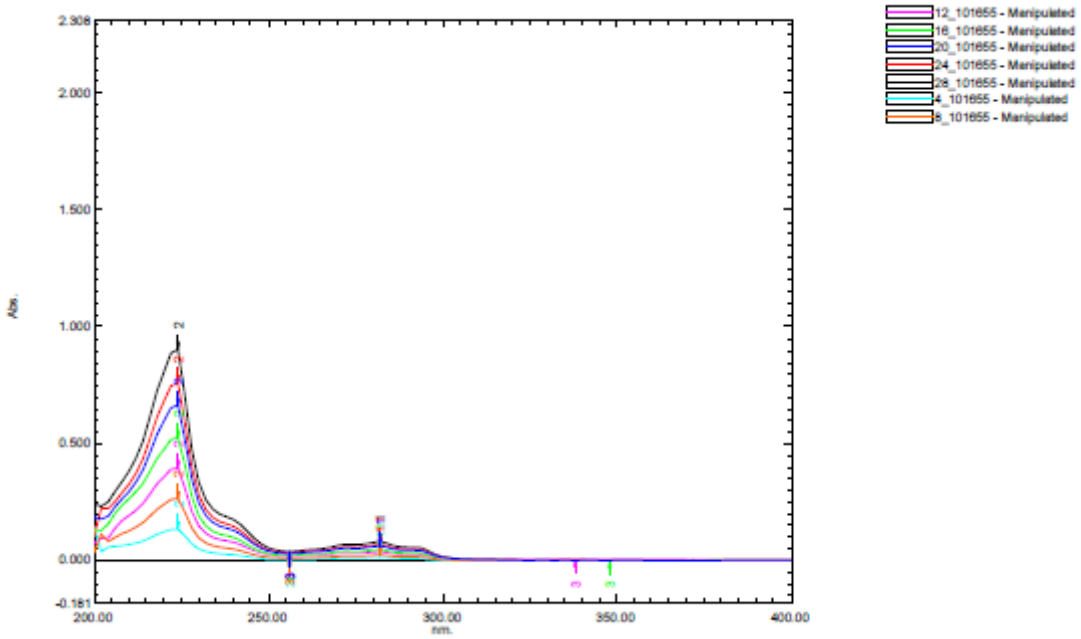


Figure No. 6: Overlay spectrum of Terbinafine

b) Precision

The term precision is defined by the ISO International Vocabulary of Basic and General Terms in Metrology (ISO-VIM) and ICH as the closeness of agreement between quantity values obtained by replicate measurements of a quantity under specified conditions. Assessing the precision implies expressing numerically the random error or the degree of

dispersion of a set of individual measurements using the standard deviation, the variance, or the coefficient of variation.

c) Repeatability

It is the concordance of a series of measurements of the same quantity when the experiments are conducted under the same conditions (analyst, apparatus, instrument, and day) in rapid succession. For this experiment, a standard solution of Terbinafine (1.6 ug/ml) was prepared and analyzed six times as per the proposed method.

d) Intermediate Precision

It is the concordance of a series of measurements of the same quantity when the experiments are conducted within the same laboratory under different conditions (analyst, apparatus, instrument, and day). A standard solution of Terbinafine (1.6μg/ml) was prepared and analyzed as per the proposed method.

Table no 2: Repeatability and intermediate precision study

S.no.	Precision	Percentage recovery of Terbinafine	% RSD
1	Repeatability	100.956±0.579	0.573
2	Intermediate Precision (Day1-Day 3)	104.833±0.893	0.851

e) Accuracy

Accuracy was determined using recovery experiments, by the determination of % mean recovery of the sample at three different levels (80-120%). At each level, three determinations were performed. Percent mean recovery was calculated as shown in **Table 3.** The accepted limits of recovery are 90% - 120% and all observed data are within the required range which indicates good recovery values and hence the accuracy of the method developed.

Table no 3: Accuracy Study of Terbinafine

Level	Amount added of Terbinafine	Percentage recovery of Terbinafine	% RSD
80%	1.2 (µg/ml)	99.581±0.693	0.696
100%	1.6 (µg/ml)	100.629±0.590	0.586
120%	2.0(µg/ml)	102.778±0.240	0.234

f) Ruggedness

Ruggedness was determined by carrying out analysis by two different analysts and the respective percentage recovery was noted and the results were indicated as % RSD.

Table no 4: Results of Ruggedness of Terbinafine

S. No.		Percentage recovery of Terbinafine	% RSD
1	Analyst 1	102.987± 0.703	0.683
2	Analyst 2	100.891±1.059	1.050

3. Limit of Detection (LOD) and Limit of Quantitation (LOQ).

The detection limit of an individual analytical procedure is the lowest amount of analyte in the sample which can be detected but not necessarily quantitated as an exact value. The Quantitation limit of an individual analytical procedure is the lowest amount of analyte in the sample which can be quantitatively determined with suitable precision and accuracy. The LOD and LOQ of the proposed method were determined by using a calibration curve:

$$\mathrm{LOD} = \frac{3.3\sigma}{S}, \qquad \mathrm{LOQ} = \frac{10\sigma}{S},$$

Where σ is the standard deviation of the response (Y-intercept) and S is the slope of the calibration curve.

Table no 5: LOD and LOQ

Drug	LOD (µg/ml)	LOQ (µg/ml)
Terbinafine at 224nm	0.030	0.091

4. Robustness

Robustness is the ability to provide accurate and precise results under a variety of conditions. To measure the extent of method robustness, the most critical parameters were interchanged while keeping the other parameters unchanged and in parallel, the chromatographic profile was observed and recorded. The studied parameter was Change in wavelength. The results for the robustness study in Table 8 indicated that the small change in the conditions did not significantly affect the determination of Terbinafine.

Table no 6: Robustness (Change in wavelength) for 1.6µg/ml

Conc.(µg/ml)	Wavelength		
	Abs1 (219nm)	Abs2 (224nm)	Abs3 (229nm)
1.6	0.438	0.527	0.285
1.6	0.44	0.527	0.285
1.6	0.444	0.534	0.291
1.6	0.453	0.527	0.289
1.6	0.448	0.539	0.28
1.6	0.449	0.529	0.285
Mean	**0.445**	**0.531**	**0.286**
SD	**0.006**	**0.005**	**0.004**
%RSD	**1.283**	**0.937**	**1.335**

CONCLUSION:

The present analytical method was validated as pet ICH Q2(R1) guideline and it meets specific acceptance criteria. It is concluded that the analytical method was specific, precise, linear, and accurate while estimating the commercial formulation without the interference of the excipients and another additive. Hence the present analytical method can be used for the

routine determination of Terbinafine in the pure pharmaceutical formulation at the minimum cost.

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REFERENCES:

1. Indian Pharmacopoeia Volume III. Published by Indian Pharmacopoeia Commission, Ghaziabad. 2010; 1750-1751.
2. S.G. Cardoso, E.S.S. Elfrides, UV spectrophotometry and nonaqueous determination of Terbinafine hydrochloride in dosage forms,J. AOAC Int. 82 (1999)830-833.
3. European Pharmacopoeia, EDQM, Council of Europe, Strasbourg, France, Edition 7.0, 2011, pp. 3047-3048.
4. British Pharmacopoeia, Her Majesty's, Stationery office, London, 2012, pp. 2112-2113.
5. United States Pharmacopeia, USP35, National Formulary-32, Rockville, USP Convention, 2012, pp. 4789-4790.
6. Basavaiah Kanakapura, Vamsi Krishna Penmatsa, Analytical methods for determination of terbinafine hydrochloride in pharmaceuticals and biological materials. Journal of Pharmaceutical Analysis. 2016;6(3):137-149.
7. International Conference on Harmonization (ICH) Guidelines Q1B: Stability Testing: Photostability Testing of New Drug Substances and Products, 1996.
8. International Conference on Harmonization (ICH) Guidelines Q1A (R2): Stability Testing of New Drug Substances and Products, 2003.
9. International Conference on Harmonization (ICH) Guidelines Q2 (R1): Validation of Analytical Procedures:Text and Methodology, 1996.
10. P.S. Jain, A.J. Chaudhari, S.A. Patel, et al., Development and validation of the UV spectrophotometric method for the determination of Terbinafine hydrochloride in bulk and in formulation, Pharm. Methods. 2(2011)198-202.
11. P.D. Goswami, Development and validation of spectrophotometric method for the determination of Terbinafine hydrochloride in bulk and in tablet dosage form,Int. J. Pharm. Technol. 5 (2013)5441-5447.
12. K.K. Patel, B.H. Marya, V.V. Kakhanis, Spectrophotometric determination and validation for Terbinafine hydrochloride in pure and in tablet dosage form.Pharm,Lett. 4 (2012)1119- 1122.
13. Iram F, Iram H, Iqbal A, Husain A (2016) Forced Degradation Studies. J Anal Pharm Res 3(6): 00073. DOI: 10.15406/japlr.2016.03.00073
14. E.M. Abdel-Moety, K.O. Kelani, A.M. Abou Al-Alamein, Spectrophotometric determination of Terbinafine in presence of its photodegradation products, Bull. Chim.Farm. 141 (2002)267-273.
15. International Conference on Harmonization (ICH) Guidelines Q1B: Stability Testing: Photostability Testing of New Drug Substances and Products, 1996.
16. International Conference on Harmonization (ICH) Guidelines Q1A (R2): Stability Testing of New Drug Substances and Products, 2003.
17. International Conference on Harmonization (ICH) Guidelines Q2 (R1): Validation of Analytical Procedures:Text and Methodology, 1996.
18. P.S. Jain, A.J. Chaudhari, S.A. Patel, et al., Development and validation of the UV spectrophotometric method for the determination of Terbinafine hydrochloride in bulk and in formulation, Pharm. Methods. 2(2011)198-202.

19. P.D. Goswami, Development and validation of spectrophotometric method for the determination of Terbinafine hydrochloride in bulk and in tablet dosage form,Int. J. Pharm. Technol. 5 (2013)5441-5447.
20. K.K. Patel, B.H. Marya, V.V. Kakhanis, Spectrophotometric determination and validation for Terbinafine hydrochloride in pure and in tablet dosage form.Pharm,Lett. 4 (2012)1119- 1122.
21. Indian Pharmacopoeia Volume III. Published by Indian Pharmacopoeia Commission, Ghaziabad. 2010; 1750-1751.
22. S.G. Cardoso, E.S.S. Elfrides, UV spectrophotometry and nonaqueous determination of Terbinafine hydrochloride in dosage forms,J. AOAC Int. 82 (1999)830-833.
23. European Pharmacopoeia, EDQM, Council of Europe, Strasbourg, France, Edition 7.0, 2011, pp. 3047-3048.
24. British Pharmacopoeia, Her Majesty's, Stationery office, London, 2012, pp. 2112-2113.
25. United States Pharmacopeia, USP35, National Formulary-32, Rockville, USP Convention, 2012, pp. 4789-4790.
26. Basavaiah Kanakapura, Vamsi Krishna Penmatsa, Analytical methods for determination of terbinafine hydrochloride in pharmaceuticals and biological materials. Journal of Pharmaceutical Analysis. 2016;6(3):137-149.