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DEVELOPMENT AND VALIDATION OF NEW UV-VISIBLE SPECTROPHOTOMETRIC METHOD FOR THE ESTIMATION OF MINOXIDIL IN BULK AND PHARMACEUTICAL DOSAGE FORM

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Abstract

Minoxidil, an oral direct-acting peripheral vasodilator, lowers high systolic and diastolic blood pressure and is also applied topically to treat androgenetic alopecia. The present investigation is to develop a simple, precise and cost-effective UV method for method development and validation of Minoxidil in bulk and pharmaceutical dosage form. The calibration curve was constructed with concentrations ranging from 20 to 60 µg/ml. The correlation coefficients were found to be 0.9993. Accuracy method was ascertained by determination of the recovery of the method at three different concentrations by standard addition method. The results were within the range of 99.57– 100.55 % and were found to be highly accurate. The parameter LOD was determined on the basis of response and slope of the regression equation. The LOD for this method was found to be 5.08µg/ml. The parameter LOQ was determined on the basis of response and slope of the regression equation. The LOQ for this method was found to be 15.38µg/ml. The samples were tested and the results of ruggedness were between the ranges of 99.75 – 99.94 %. These values were found to be within the limits. In the present investigation, simple and sensitive UV spectrophotometric methods were developed for the quantitative estimation of Minoxidil in bulk drug and pharmaceutical formulations. In addition to positive requirements of these analytical methods, the striking advantages of all the presently developed methods were economical. The methods were validated in terms of linearity, accuracy, precision, ruggedness and robustness and used for the outline determination of Minoxidil in bulk drug and in pharmaceutical formulations.

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Introduction

Minoxidil, an oral direct-acting peripheral vasodilator, lowers high systolic and diastolic blood pressure and is also applied topically to treat androgenetic alopecia [1]. Minoxidil is a crystalline powder with molecular formula C9H15N5O (Molecular weight 209.251 g/mol). It is chemically 2,4-Diamino-6-piperidinopyrimidine-3-oxide. It acts by causing vascular smooth muscle cells adenosine triphosphate-sensitive potassium channels to open and through the activation of extracellular signal-regulated kinase and the prevention of cell death. Minoxidil is believed to enhance the life of human follicle dermal papillary cells or hair cells. In the present study, the authors have proposed new spectrophotometric techniques for the assay of Minoxidil in topical solution and validated as per ICH guidelines. Analytical methods such as spectrophotometry [2, 3], liquid chromatography and mass spectroscopy were developed for the determination of Minoxidil in pharmaceutical dosage forms and biological fluids.

According to the literature survey it was found that few analytical methods were reported for the estimation of Minoxidil by using UV spectroscopy. The other methods were also proposed for its determination includes HPLC, RP-HPLC, and electrochemical method [4, 5]. The present investigation is to develop a simple, precise and cost-effective UV method for method development and validation of Minoxidil in bulk and pharmaceutical dosage form.

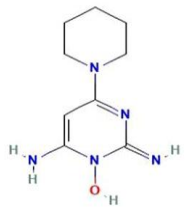

Figure 1: Structure of Minoxidil

Materials and Methods

Minoxidil was obtained as a gift sample from The Madras Pharmaceuticals, Chennai and marketed formulations of Minoxidil, Minoxiheart (5 mg) were procured from Steris Healthcare Private Limited, Mumbai. Minpro (10 mg) were procured from Jeerima BioCare Private Limited, Mumbai. All the chemicals use are AR Grade. UV Visible spectrophotometer (Systronics 2202) was employed with spectral bandwidth of 1 nm attach with computer loaded PC software (UV probe) version 2.31.

Preparation of standard primary drug stock solutions

Standard drug stock solution of Minoxidil (1mg/ml or 1000 µg/ml) was prepared by dissolving equivalent to 25mg of Minoxidil in 25 ml of volumetric flask containing 25 ml of solvent as Ethanol [6].

Preparation of standard secondary drug stock solution

From the above standard primary drug solution, 1ml of stock solution was withdrawn and diluted with 100ml of 0.1N Ethanol to produce 100µg/ml concentration.

Preparation of linearity working standard solution

Linearity standard solutions were prepared from secondary standard stock drug solution. Pipette out 2ml, 3ml, 4ml, 5ml, & 6ml, from secondary stock solution and transfer them in to 10 ml volumetric flasks and further dilute it to 100ml with primary diluent to get a final linearity concentration of 20µg/ml to 60µg/ml of Minoxidil [7].

Preparation of standard solution preparation

A working standard solution concentration having 100 µg/ml of Minoxidil was prepared from the above secondary standard stock solution.

Preparation of sample solution

Five tablets of Minoxidil were weighed and finely powdered. An accurately weighed quantity of the tablet powder equivalent to approximately 25 mg of Minoxidil was transferred to a 25 ml standard flask. This was then diluted with 25 ml of 0.1 N ethanol to obtain a concentration of 100 µg/ml. From this 2ml, 3ml ,4ml, 5ml& 6ml of sample solution was taken and diluted with 10ml of 0.1N Ethanol to give 20, 30, 40, 50 & 60µg/ml concentration. The solution was filtered and absorbance value of sample solutions was recorded at 280 nm.

The working standard stock solution was prepared and scanned the spectrum by UV spectrophotometer range between 200-400 nm. After careful observation of spectrum, the λ max was obtained as 286nm and spectrum showed at Figure 2.

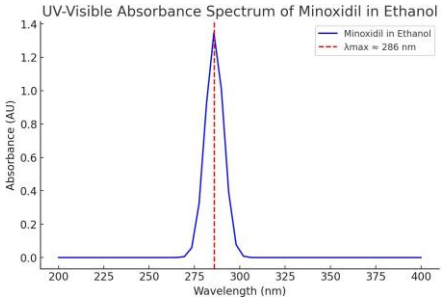

Fig.2: Absorption spectrum of Minoxidil from 200nm-400nm

Results and Discussion

Validation is defined as the establishing evidence which provide high degree of assurance that a specific process will consistently produce a product meeting its determined specification quality characteristics [8]. The following parameters used for validation studies are

Linearity

The calibration curve was constructed with concentrations ranging from 20 to 60 µg/ml. The absorbance of the drug was considered for plotting the graph. The linearity was evaluated by linear regression analysis, which was calculated by the least square regression method. The linearity data was showed in Figure 2. The correlation coefficients were found to be 0.9993.

Table 1. Linearity data for minoxidil

S.No	Concentration (ug/ml)	Absorbance
1	20	0.14
2	30	0.27
3	40	0.40
4	50	0.53
5	60	0.66

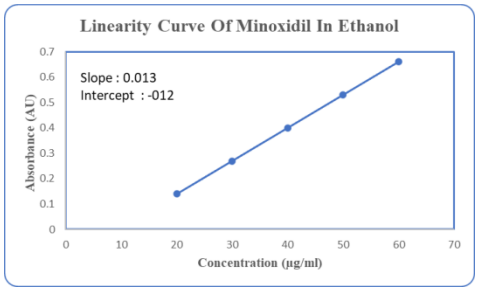

Fig.3. Linearity Curve of Minoxidil in Ethanol

Accuracy

Accuracy method was ascertained by determination of the recovery of the method at three different concentrations by standard addition method [9]. It was performed by adding 80%, 100%, 120% of pure standard drug of Minoxidil to previously analyzed tablet powder sample and mixtures were reanalyzed by the proposed method (Table 2). The results were within the range of 99.57–100.55 % and were found to be highly accurate.

Table 2. Recovery studies

Tablet Sample	Level of Recovery %	Amount Present (mg/tab)	Amount Standard (mg)	Total amount Recovered (mg)	Percentage Recovery
T1	80	5	4	9.10	101.11
		5	4	9.35	103.88
		5	4	8.99	99.89
	100	5	5	10.30	103.00
		5	5	11.08	110.80
		5	5	9.89	98.90
	120	5	6	11.45	104.09
		5	6	12.08	109.82
		5	6	11.07	100.64

Precision

Precision may be considered at three levels: repeatability, intermediate precision and reproducibility [10]. Intraday and Inter day precision were evaluated by determining the corresponding responses three times on the same day and on 3 different days for solution containing 10µg ml of Minoxidil. The precision of the proposed method i.e. the intra and inter-day variations in the absorbance of the drug solutions was calculated in terms of % RSD and the results are presented in Table 3.

Table 3. Precision Studies

Concentration (µg/ml)	Inter-day Absorbance Mean ± SD	% RSD	Intra-day Absorbance Mean ± SD	%RSD
20	0.055 ± 0.0015	1.206	0.055 ± 0.0018	1.886
30	0.085 ± 0.0021	1.184	0.085 ± 0.0024	1.554
40	0.117±0.0029	0.391	0.117±0.0030	1.270
50	0.139 ± 0.0035	0.084	0.139 ± 0.0037	1.176
60	0.167 ± 0.0043	0.813	0.167 ± 0.0045	0.109

Acceptance criteria: %RSD of 5 replicate preparations of assay should be not more than 2%.

Table 4: Statistical validation for precision

Component	Mean*	S.D	C.O.V.	S.E
Intra-day	**99.77**	**0.00286**	**0.0028666**	**0.001279**
Inter-day	**99.50**	**0.00308**	**0.00309547**	**0.001377**

Limit of Detection (LOD)

The parameter LOD was determined on the basis of response and slope of the regression equation. The LOD for this method was found to be 5.08µg/ml.

Limit of Quantification (LOQ)

The parameter LOQ was determined on the basis of response and slope of the regression equation. The LOQ for this method was found to be 15.38µg/ml.

Ruggedness

Minoxidil tablet equivalent to 5 mg was weighed and dissolved in a 10 ml volumetric flask containing solvent (5.5 ml), sonicated for few mins and the final volume was made with remaining solvent. The samples were tested and the results are recorded and, the results of ruggedness were shown in the table 5. The values were between the ranges of 99.75 – 99.94 %. These values were found to be within the limits.

Table 5. Ruggedness results for Minoxidil at 286 nm

Sample	Label claim (mg)	Analyst I		Analyst II	
		Amount found (mg)	Recovery ± SD** (%)	Amount found (mg)	Recovery ± SD** (%)
MinoxiHeart	**5mg**	4.9875	99.75 ± 0.09	4.9895	99.79± 0.11

Determination of Minoxidil Tablets

UV Spectrophotometric method developed is sensitive and specific for the estimation of Minoxidil [11, 12]. Also the method is validated for different parameters, hence has been applied for the Limitation of drug in pharmaceutical dosage forms [13]. Tablets (containing were evaluated for the amount of Minoxidil present in the formulation.

Table 6. Assay of Minoxidil Tablets

Tablet	Concentration	Amount present (mg/tab)	Amount found (mg/tab)	% of drug found
T1	40	10	39.40	98.5
	70	10	68.10	97.28
	80	10	80.20	100.25

*Average of six determination Acceptance criteria for % Recovery should be between 90 to 110%. Since the % R.S.D of Percent recovery was found to be below 2%, the assay parameter was passed.

Conclusion

The UV spectroscopy method developed for the determination and validation of Minoxidil was found to be accurate, precise, and reliable. The method demonstrated excellent linearity, sensitivity, and reproducibility within the selected concentration range. The validation parameters, including accuracy, precision, specificity, and robustness, met the required regulatory standards, confirming the method's suitability for routine analysis of Minoxidil in pharmaceutical formulations [14]. This study establishes UV spectroscopy as a simple and cost-effective technique for the quantification of Minoxidil, ensuring quality control and compliance with analytical requirements. In the present investigation, simple and sensitive UV spectrophotometric methods were developed for the quantitative estimation of Minoxidil in bulk drug and pharmaceutical formulations. In addition to positive requirements of these analytical methods, the striking advantages of all the presently developed methods were economical. The methods were validated in terms of linearity, accuracy, precision, ruggedness and robustness and used for the outline determination of Minoxidil in bulk drug and in pharmaceutical formulations.

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Conflict of Interest

The authors declare no conflict of interest.

Author Contribution

Concept: Y. Kesava, design: Y. Kesava, data collection: Amarjeet Kumar, Kusuma, Ankitha, Paul Elisha, Richa Halder, analysis: Dr. D.Jothieswari, writing: Amarjeet Kumar, Kusuma, Ankitha, Paul Elisha, Richa Halder.

References

1. Zahid A Zaheer, et al. (2012). UV-Spectrophotometric Determination of Minoxidil and its Application to the Assay in Pharmaceutical Dosage Forms. Der Pharma Chemica, Vol. 4 (1), 568-573.
2. Vijayamma G, et al. (2015). Analytical Method Development and Validation of Minoxidil in Pharmaceutical Dosage Forms by UV Spectrophotometry. International Journal of Innovative Pharmaceutical Research, Vol. 6 (1), 464-467.
3. Maryam Bordbar, et al. (2009). Simultaneous Spectrophotometric Determination of Minoxidil & Tretinoin by the H-Point Standard Addition. Chemical Papers-Slovak Academy of Sciences. Vol. 63 (3), 336-344.
4. Nivedita Patel & Dhananjay B Meshram. (2015). Development and Validation of Analytical Method for Simultaneous Determination of Minoxidil and Finasteride in Pharmaceutical Dosage Form by RP-HPLC Method. International Journal of Pharmaceutical Science and Research. Vol. 6 (11), 4882-4885.

5. Patel PD, et al. (2015). Development and Validation of Stability Indicating Method for Minoxidil and Finasteride in its Pharmaceutical Dosage Form. International Journal for Pharmaceutical Research Scholars. Vol. 4 (2), 221-238.
6. M. Sudhakar, et al. (2015). Analytical Method Development and Validation for Simultaneous Estimation of Finasteride and Minoxidil in Pharmaceutical Dosage Form by RP-HPLC Method. International Journal of Pharma Sciences. Vol. 5 (2), 954-959.
7. R. A. de Sousa, et al. (2005). Fast Determination of Minoxidil by Photometric Flow Titration. ECLETICA Quimica Sao Paulo. Vol. 30 (3), 79-84.
8. Akhilesh Sharma, et al. (2016). Stability Indicating Simultaneous Validation of Azelaic acid, Minoxidil and Tretinoin with Forced Degradation Behavior Study by RP-HPLC in Pharmaceutical Dosage Form. European Journal of Biomedical and Pharmaceutical Sciences. Vol. 3 (11), 272-281.
9. Hemant K Gaidhane, et al. (2011). Development and Validation of Stability Indicating HPLC Method for the Estimation of Minoxidil and Related Substances in Topical Formulation. Journal of Pharmacy Research. Vol. 4 (12), 4481-4484.
10. A. Aboul-Kheir, et al. (2012). Bromometric Analysis of Lamotrigine, Minoxidil and Cefixime. Asian Journal of Pharmaceutical Analysis. Vol. 2 (1), 22-28.
11. S. Siddiraju, et al. (2014). Stability Indicating RPHPLC Method Development and Validation for the Simultaneous Determination of Aminexil and Minoxidil in Pharmaceutical dosage Form. Annales Pharmaceutiques francaises. Vol. 73 (2).
12. A. Aboul-Kheir, et al. (2012). Spectrophotometric Estimation of Lamotrigine and Minoxidil in Bulk and Dosage Forms. Research Journal of Pharmacy and Technology. Vol. 5 (5), 697-708.
13. P. A. Asmus, et al. (1984). Determination of Minoxidil in Bulk Drug and Pharmaceutical Formulations by Ion-Pairing HPLC. Journal of Pharmaceutical Sciences.
14. A Ruiz-Medina, et al. (1999). Integrated Flow Injection- Solid Phase Spectrophotometric Determination of Minoxidil. Vol. 50 (2), 277-282.