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Validated spectrophotometric method for quantitative determination of Artemether in pharmaceutical formulation

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ABSTRACT

The simple, accurate and precise UV-Spectrophotometric method has been developed and validated for quantitative determination of artemether in capsules. The method employed lN HCl as a solvent and used to derivatize a drug. The proposed method obeyed the Beer's law in the concentration range of 5-40 µg/ml. The linear regression showed good linear relationship with r^2 = 0.998, slope and intercept were 0.0182 and 0.006622 respectively. Method was validated statistically where SD and RSD were found to be satisfactorily low. Percentage recovery of the drug for the proposed method was found in the range of 99.5-100.86% indicating no interference of the capsule excipients. The results of the capsule analysis were validated with respect to accuracy, precision and recovery studies which were found to be satisfactory. LOD and LOQ for artemether were found to be 2.30 µg/ml and 4.08 µg/ml respectively. The utility of the developed method has been demonstrated by analysis of commercial formulation containing this drug.

Key Words: Artemether, UV Spectrophotometry, Derivatization, 1N HCl, Distilled Water.

INTRODUCTION

Chemically, Artemether (ART), also called dihydroartemisinin methyl ether, is a synthetic derivative of artemisinin, widely used in malaria treatment in endemic areas [1]. This drug can be administered as an oily solution by intramuscular injection or in capsules orally. ART is active against the plasmodium genera that cause malaria. The key structure characteristic appears to be a "trioxane" consisting of endoparoxide and doxepin oxygen's which are simpler class of 3-aryl trioxane [2]. Endoparoxide moiety is required for antimalarial activity whereas substitution on lactone carbonyl group markedly increase potency. Its in vivo potency is 10 – 100 folds greater than other antimalarial drugs [3]. Artemether-lumefantrine combination is the first fixed dose oral combination of an artemisinin derivative with a second unrelated antimalarial component [4].

Literature survey reveals that few analytical methods like HPLC [5], stability indicating [6], GC-MS [7], HPTLC [8] and UV spectrophotometry [9-11] are reported. As each method suffers from its own limitations, so here an attempt has been made to develope a new UV spectrophotometric method for estimation of ART in pharmaceutical formulation with accuracy, simplicity, precision and economy.

In the present study 1N HCl was used to derivatize a drug at 80°C for 20 minutes and water was used as a diluting solvent. ART was found to be freely soluble in 1N HCl at 80°C and hence it was selected for derivatization and solubilization of drug. The proposed UV spectrophotometric method is a simple, specific, rapid, and accurate for quantitative estimation of ART. The results obtained have been statistically validated in accordance with the ICH guidelines [12] and can be effectively used for the determination of ART in bulk and pharmaceutical formulations.

MATERIALS AND METHODS

2.1 Chemicals and Reagents

A standard gift sample of ART was provided by Shreya Life Sciences Ltd, Aurangabad. Commercially available formulations were procured from local market. Spectroscopic Grade HCl was purchase from LOBA Chemie Pvt Ltd., Mumbai. Distilled water was used throughout the study and 1N HCl was used for derivatization of drug.

2.2 Instrumentation

UV- Visible double-beam spectrophotometer, Shimadzu model 1700 with spectral bandwidth of 1 nm, wavelength accuracy ± 0.3 nm and a pair of 10-mm matched quartz cells was used. All the weighing was done on electronic balance.

2.3 Preparation of Standard Solution:

Accurately weighed 10 mg of ART was transferred to a 100 ml volumetric flask. To it add 25 ml of 1 N HCl and this solution was heated on the water bath for 20 minutes at temperature 80 ± 2ºC. The solution was allowed to cool at room temperature and volume was then made up to the mark with distilled water to get concentration of 100 µg/ml and used it as a stock solution.

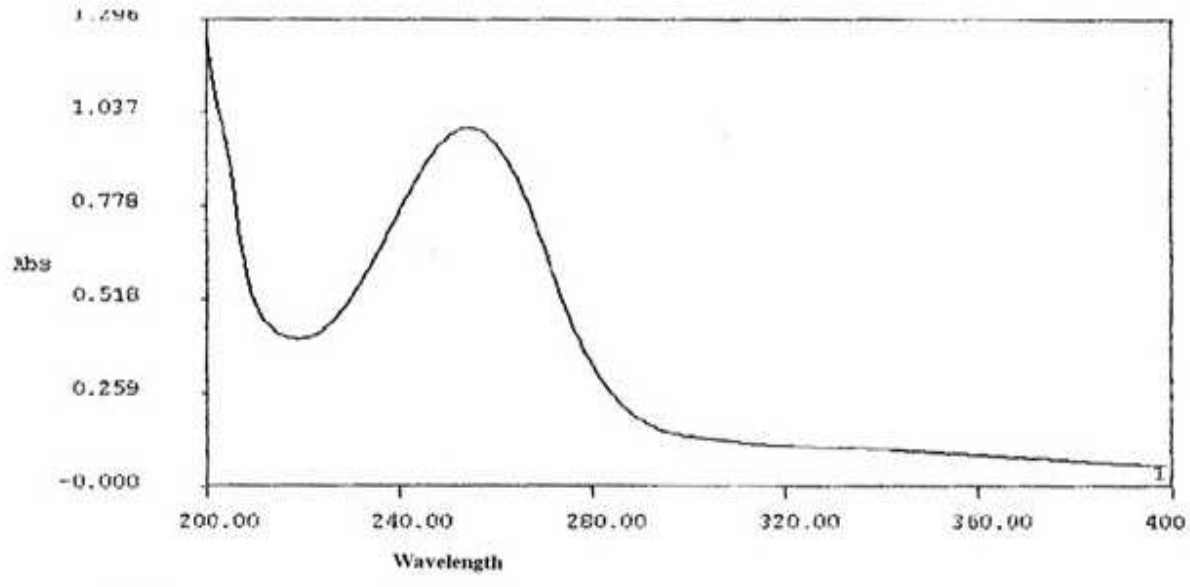

Fig. 1 : Absorption maxima of ART(20µg/ml) against reagent blank

2.4 Determination of Absorbance maxima:

The stock solution was further diluted with distilled water to get concentration of 20µg/ml. This solution was then scanned in the range of 200 – 400 nm where distilled water was used as a blank. The wavelength of maximum absorbance of ART was found at 254 nm as shown in **Fig. 1**

2.5 Method

Suitable aliquots of the stock solution of ART (0.5 – 4 ml) were taken in 10 ml volumetric flasks. Flasks were shaken for few minutes and volume was then made up to the mark with distilled water to prepare a series of standard solutions containing 5-40 µg/ml in the concentration range. Absorbance of the complex was measured at 254 nm against blank. Blank was prepared by heating 2.5 ml 1N HCl in the same condition and diluting up to the 10 ml with distilled water. Then calibration curve was plotted for ART in the concentration range of 5-40 µg/ml at 254 nm as shown in **Fig. 2**.

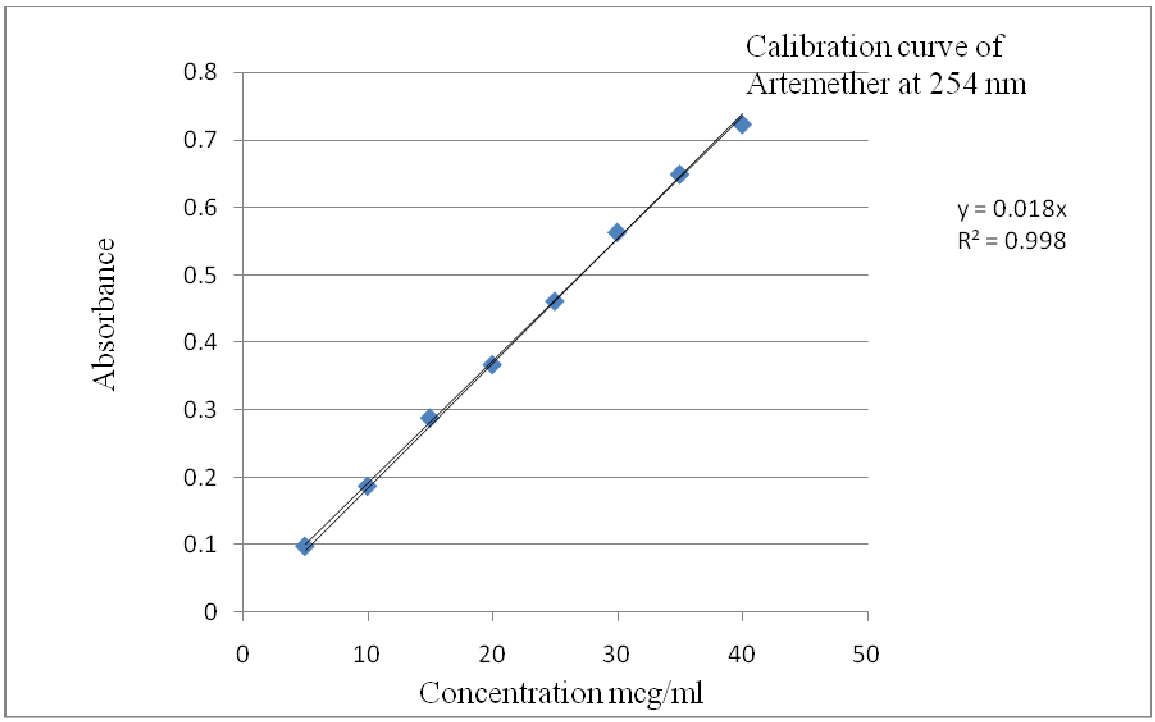

Fig. 2: Calibration curve of ART at 254nm

Table No.1: Optical characteristics and Other Parameters

Parameters	UV Spectrophotometric Method
λ max (nm)	254
Beer's law limits (µg/ml)	5 – 40
Molar absoptivity ($l.mol^{-1}cm^{-1}$)	5.594×10^4
Correlation coefficient (r^2)	0.998
Slope (m)	0.0182
Intercept (c)	0.006622
LOD (µg/ml)	2.30
LOQ (µg/ml)	4.08

2.6 Estimation of ART in capsules

For the estimation of drug in the commercial formulation, twenty capsules were weighed and the capsule powder was removed. The weight of empty capsule shells was then recorded. Difference

in the weight of filled capsules and empty capsule shells was calculated to know the weight of powder present in twenty capsules. Average weight was then calculated. The quantity of powder equivalent to about 10 mg of ART was transferred to a 100 ml volumetric flask and mixed with 25 ml 1N HCl and solution was sonicated for 10 minutes. Then solution was heated on the water bath for 20 minutes at temperature 80 ± 2ºC. The solution was allowed to cool at room temperature after heating and then diluted up to the mark with distilled water. The solution was filtered through Whatmann filter paper No. 42. Reference standard of ART was also treated in the same way at each concentration and absorbance was noted at 254 nm against blank. The result of analysis of ART from capsule formulation is shown in **Table No. 2.**

Table No.2: Result of Analysis of Capsules in marketed formulation

Brand Name	Label Claim (mg/capsule)	Amount of drug estimated* (mg/capsule)	% of label claim Estimated ± S.D*
Larither	40 mg	39.17	97.93 ± 0.98

** Mean of six determinations.*

3. Method Validation

The proposed methods were validated as per ICH guidelines.

3.1 Accuracy

To ascertain the accuracy of the proposed method, recovery studies were carried out by standard addition method at three different levels (80%, 100% & 120%). The results of recovery studies, expressed as percent recovery, were satisfactory and are presented in **Table No.3.**

Table No. 3: Result of recovery studies

Sr.No.	Level of Recovery (%)	Fixed amount added (µg)	Amount added (µg)	Amount Estimated (µg)	Recovery* (%)	(±) S.D.*	R.S.D.*
1	80	20	16	36	100	0.35	0.0097
2	100	20	20	39.8	99.5	0.06	0.0015
3	120	20	24	44.38	100.86	0.48	0.0108

**Mean of six determinations,*

3.2 Precision

The reproducibility of the proposed methods was determined by analyzing capsules at different time intervals on same day (Intra-day assay precision) and on three different days (Inter-day assay precision). Coefficient of variance for intra-day and inter-day assay precision was found to be 0.218 and 0.221 respectively.

RESULTS AND DISCUSSION

The assay values for capsule were in the range of 98.05-99.00%. The optical characteristics and other parameters are shown in **Table No.1.** The recovery study was carried out by standard addition method. The results of recovery study for the drug was in the range of 99.5-100.86% as shown in **Table No.3**. Statistical analysis was performed on the results of nine determinations at three different concentration levels. The Standard deviation and relative standard deviation was used to check the accuracy of method. Intraday and interday assay precision was used to check the precision of method.

CONCLUSION

Based on the results obtained, it can be concluded that the proposed UV spectrophotometric method for determination of ART is rapid, economical, accurate and precise. The utility of the developed method has been demonstrated by analysis of capsule formulation. Hence, the proposed method can be used for quantitative determination of pharmaceutical formulation containing this ingredient.

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REFERENCES

[1] I.C. Cesar, G.A., Pianetti, *Braz. J. Pharm.Sci.*, **2009,** 45, 737-742.
[2] J.H. Block, J.M. Beale, Jr., Wilsone and Gisvold's, Text Book of Organic Medicinal and Pharmaceutical Chemistry, eleventh edition, **2004** P. 294-295.
[3] J.G.Hardman, L.E.Limbird, Goodman and Gilman's The Pharmacological Basis of Therapeutics, 10th international edition, 1072.
[4] T.M. Davis, H.A. Karunanejeewa, K.F. Illet, *Medical Journal*, **2005**, 182, 181-185.
[5] C.G. Thomas, S.A. Ward, G. Edwards, *J. Chromatogr.*, **1992,** 583:1, 131- 136.
[6] A. Shrivastava, R. Issarani, B.P. Nagori, *J. Young Pharmacists*, Volume 2 (1), Jan-Mar **2010**, 79-84.
[7] S.S. Mohamed, S.A. Khalid, S.A. Ward, T S Wan et.al, *J Chrom B Biomed Sci Appl.*,**1999**, 731: 2, 251-260.
[8] N.G. Tayade and M.S. Nagarsenker, *J Pharm Biomed Anal*, 43: 3, **2007**, 839-844.
[9] A.Shrivastava, BP Nagori, P.Saini, R.Issarani and SS Gaur**,** *Asian J. Research Chem.*, 1:1, July- Sept. **2008**,19-21
[10] O.A. Adegoke, O.S. Idowu, O.P. Daramola and O.S. Ogunsanya, *Acta Pharma. Sci*, **2010,** 52, 269-280.
[11] The International Pharmacopoeia, World Health Organization Geneva, Third Edition, Volume 5, **2003**, 187-190.
[12] International Conference on Harmonization (ICH), Harmonized Tripartite Guidelines on 'Validation of Analytical Procedures: Methodology', Operational from June **1997**, The European Agency for the Evaluation of Medicinal products, Human Medicines Evaluation Unit.