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Development of Four Simple UV-Spectrophotometry Methods for Estimation of Garenoxacin Mesylate in Bulk Material and in Pharmaceutical Formulation

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ABSTRACT

Garenoxacin Mesylate is used in the treatment of certain respiratory tract infections Urinary tract infection. Rapid, Simple, cheap and accurate UV- Spectrophotometry methods have been developed using distilled water as solvent to determine Garenoxacin Mesylate in bulk and in tablets. "Method A" is Zero Order UV- Spectrophotometry using absorbance, "Method B" is zero order UV- spectrophotometry using AUC technique, Method C is First Order Derivative UV- spectrophotometry using amplitude and 'Method D' is First Order Derivative UV-spectrophotometry using AUC. The proposed methods have revealed best results in terms of linearity, accuracy, precision and LOD and LOQ for bulk drug and in pharmaceutical formulation. In all the proposed methods, Garenoxacin Mesylate followed linearity in the concentration range of 3 - 18 µg/mL with (r^2 > 0.999). The amounts determined by all the methods were found to be in agreement with label claimed.

Keywords: Garenoxacin Mesylate; UV- Spectrophotometry; First Order Derivative Spectrophotometry; Area under Curve

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INTRODUCTION

Garenoxacin Mesylate (GRN) is 1-Cyclopropyl-8-(difluoromethoxy)-7-[(1*R*)-1-methyl- 2,3-dihydro-1*H*-isoindol-5-yl]-4-oxo-1,4-dihydroquinoline-3-carboxylic acid methanesulfonate [1]. The chemical structure of Garenoxacin Mesylate is shown in Figure 1.

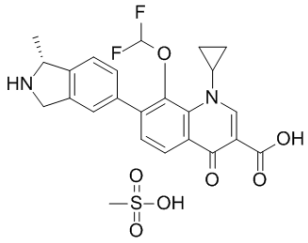

Figure 1: Chemical Structure of Garenoxacin Mesylate

Garenoxacin Mesylate (GRN) is used in the treatment of certain respiratory tract infections (RTIs), Urinary tract infection (UTI), otorhinolaryngological infections and Penicillin and fluoroquinolone-resistant Streptococcus pneumonia. [2].

A detail literature survey for GRN revealed that, few analytical methods like RP-HPLC have been reported for estimation of GRN in Pharmaceutical dosage form [3, 4] and biological fluid [5]. Also, a spectroscopic method [6] has been established.

To our understanding no methods have been found in literature for quantification of GRN in bulk and in tablet formulation using distilled water as solvent.

Therefore, our endeavor is to establish four simple Zero Order and first order Derivate UV- Spectrophotometry methods using different techniques.

The Area under Curve (AUC) technique involves calculations of included value of area with respect to the wavelength between the two selected wavelengths $\lambda 1$ and $\lambda 2$. Selection of wavelength range is on the basis of recurring observations so as to acquire the linearity between AUC and concentration [7]. Further, methods were validated as per ICH guidelines [8].

MATERIALS AND METHODS

2.1 Materials

Garenoxacin Mesylate was obtained as gift sample from Alkem Pharmaceuticals Ltd, Mumbai, India. Distilled water used as solvent was prepared in laboratory.

2.2 Instrument

A double beam UV-VIS spectrophotometer (UV-2450, Shimadzu, Japan) connected to computer loaded with spectra manager software UV Probe 2.21 with 10 mm quartz cells was used. The spectra were obtained with the instrumental parameters as follows: wavelength range: 400-200 nm; scan speed: medium; sampling interval: 10 nm; spectral slit width: 1 nm. An electronic balance (Model Shimadzu AUX 120) was used for weighing purpose.

2.3 Preparation of Stock Standard Solution

The stock standard solution of GRN was prepared by dissolving accurately weighed 10 mg in 100 mL of water to obtain a concentration of 100 µg/mL.

2.4 Methods A (Zero Order Spectrophotometry) and Method B (Zero order Spectrophotometry–AUC)

From the stock standard solution, an appropriate volumes in the range of 0.3 – 1.8 mL were transferred into series of 10 mL volumetric flask and volume was made up to the mark to obtain concentration in the range of 3-18 µg/mL. In Method A, absorbance was recorded at 274 nm while in Method B, AUC was selected in the wavelength range of 256.50 – 286 nm. The calibration curves for GRN were constructed by plotting concentration *versus* absorbance and AUC in Method A and B, respectively.

2.5 Methods C (First order derivative –UV Spectrophotometry and D (First order derivative –UV Spectrophotometry-AUC)

For Method C and D, spectra of above prepared solutions in the range of 3 - 18 µg/ mL were derivatized into first order using software UV-Probe 2.21 with delta lambda 10 and scaling factor 10. In Method C, the amplitude was recorded at 286 nm whereas in Method D, AUC of the derivative spectrum was considered at 277.50 - 300 nm. The calibration curves were constructed by plotting concentration versus amplitude in Method C, while in Method D, it was studied as concentration versus AUC of First order Derivative spectra between selected wavelengths.

The UV- and derivative- spectrum and selection of wavelengths selection is shown in **Figure 2**.

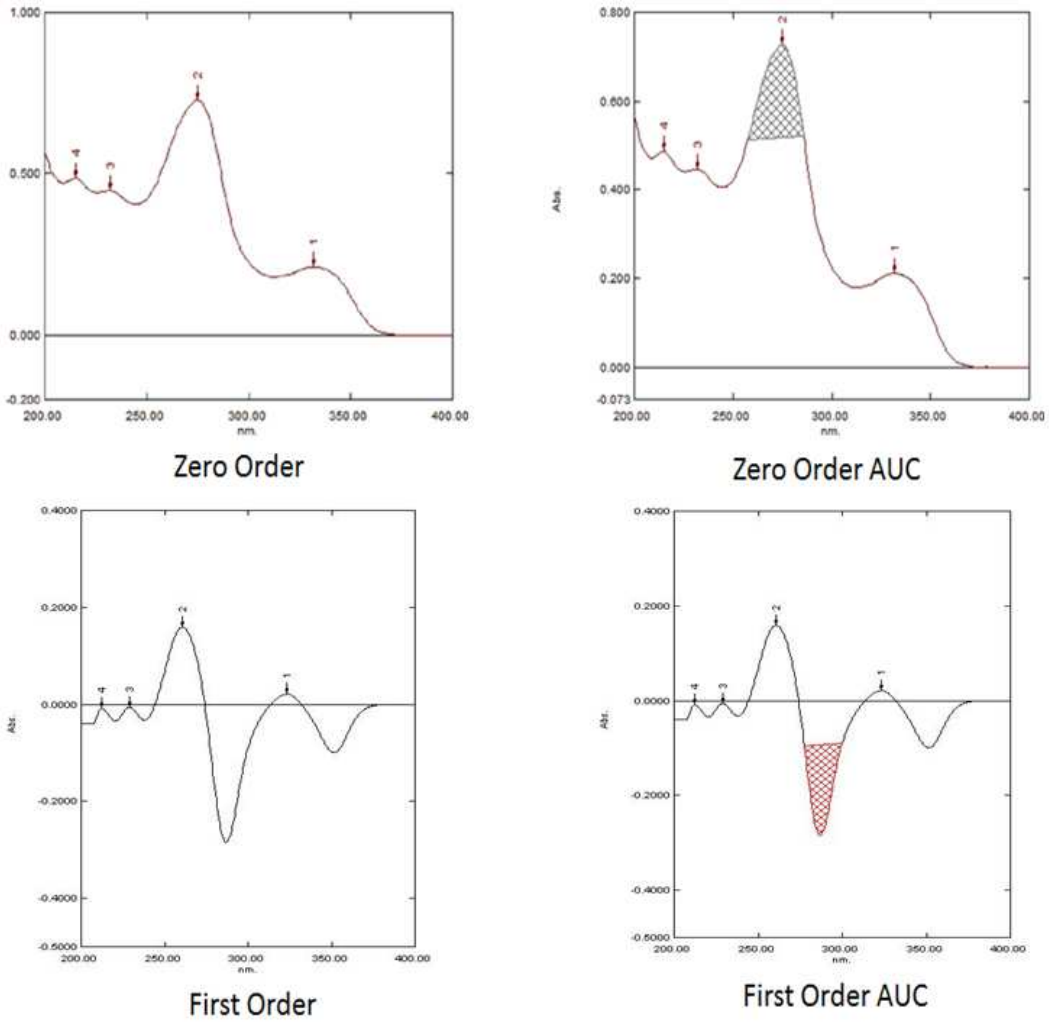

Figure 2: Zero order spectrum, First order derivative spectrum of GRN and AUC between selected wavelengths

2.6 Preparation of Sample Solution

Ten Garenoxacin Mesylate tablets (label claim 200 mg) were weighed, transferred to a clean dry mortar, and grounded into a fine powder using a pestle. Tablet powder equivalent 100 mg of GRN was transferred to a 100 mL volumetric flask containing 50 mL distilled water. It was shaken manually for 20 min and then volume was made up to the mark with distilled water and filtered through Whatmann filter paper (No. 41). From the filtrate, a correct volume was taken and diluted with distilled water to get the final concentration of 9 µg/mL. The responses were measured and concentration in the sample was determined from respective linearity equations.

3. Validation of method

The developed Method A, B, C and D were validated as per ICH guidelines.

3.1 Accuracy/ Recovery studies

The accuracy of all the methods was evaluated by recovery experiment. To the pre-analyzed sample solutions (6 µg/mL), known amounts of stock standard solutions were added at different levels, i.e. 80 %, 100 %, and 120 %. The solutions were re-analyzed by the anticipated methods. The experiments were repeated for three times at each level for each method.

3.2 Precision
Precision of the methods was studied as intra-day and inter-day variations. In all the methods precision was determined by analyzing the 6, 9 and 12 µg/mL of GRN solutions as intra-day and inter-day variations.

3.3 Sensitivity
The sensitivity of measurement of garenoxacin mesylate by the use of the proposed methods was estimated in terms of the limit of quantification (LOQ) and the limit of detection (LOD). The LOQ and LOD were calculated using equation LOD = 3.3*N/B and LOQ = 10*N/B, Where 'N' is the standard deviation of the absorbance, amplitude and peak areas of the GRN (n=3), taken as a measure of noise, and 'B' is the slope of the corresponding calibration curve.

3.4 Repeatability
Repeatability was determined by analyzing 9 µg/mL concentration of GRN solutions for six times for all methods.

3.5 Ruggedness
Ruggedness of the proposed methods was determined for 9 µg/mL of garenoxacin mesylate by analysis of aliquots from a homogenous slot by two different analysts using the same operational and environmental conditions for all methods.The results are in acceptable range that is % RSD values <2 for all the methods.

RESULT AND DISCUSSION

In distilled water, GRN obeyed linearity in the concentration range of 03 – 18 µg/mL. The wavelength for maximum absorbance/wavelength range and correlation coefficient for the all methods are given in **Table 1**.

Table 1: Optical Characteristics of GRN

Parameters	Method A	Method B	Method C	Method D
Beer-Lambert's range (µg/mL)	03-18	03-18	03-18	03-18
λ max (nm)/Wavelength range (nm)	274	256.50-286	286	277.50 - 300
Slope	0.0617	0.3444	0.0617	0.2267
Intercept	0.0053	0.0265	0.0053	0.1672
Correlation coefficient	0.997	0.997	0.998	0.997

Tablet formulation of GRN was analyzed. The amounts of GRN determined by all the methods are given in **Table 2**.

Table 2: Analysis of Tablet Formulation

Methods	% Amount found	%RSD
A	98.81 ± 1.35	1.37
B	98.15 ± 0.32	0.33
C	99.39 ± 1.53	1.54
D	98.29 ± 0.66	0.68

In all the proposed analytical methods for GRN precision studies were performed as inter-day and intra-day variations and results were studied as % relative standard deviation which was found to be less than 2). The repeatability studies in all the four methods were performed and results were determined as %RSD which were found to be less than 2. The accuracy of all the methods was determined by calculating mean percentage recovery. It was determined at 80, 100 and 120 % level and calculated in terms of % RSD value which was found to be less than 2 in all the developed methods. The ruggedness of the methods was studied by two different analysts using the same operational and environmental conditions.

The results of validation parameters are summarized in **Table 3**

Table 3: Validation Parameters

Parameters		Methods A	B	C	D
Accuracy (% Recovery)	**80 %**	98.61	98.90	98.04	100.17
	100%	99.26	99.13	98.10	99.67
	120%	98.19	99.13	98.85	98.82
Precision (% RSD)	**Intra-day (n=3)**	0.72	0.48	0.72	1.06
	Inter-day	0.85	0.40	1.03	1.48
Repeatability (% RSD)		1.22	0.17	1.22	1.84
Ruggedness (% RSD)	**Analyst I**	0.29	0.58	0.29	1.12
	Analyst II	1.11	0.38	1.11	0.59
Limit of Detection		0.04	0.10	0.04	0.11
Limit of Quantification		0.14	0.32	0.14	0.35

CONCLUSION

Four UV- Spectrophotometry methods have been developed for the estimation of GRN in bulk and in tablets are simple, economical and repeatable. All these four methods have been validated as per ICH guidelines and found to be accurate, précised, rugged and sensitive. The proposed methods can routinely be used for analysis of GRN in bulk and in pharmaceutical formulation.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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REFERENCES

[1] Martindale, 'The complete drug reference', 36th edn, Pharmaceutical press, USA, **2009**, pp. 281
[2] A. Liebetrau, A.C. Rodloff, J. Behra-Miellet and L. Dubreuil, *Antimicrob. Agents Chemother.*, **2003**, 47(11), 3667
[3] A. Unnisa, S.S. Ali, K. Siva Chaithanya, *World J. Pharmacy Pharm. Sci*., **2014**, 3 (10), 1767
[4] G. Supriya, R. Kandisa, B.V.V. Ravi Kumar, *J. Biotech. Biomater*., **2012**, 2 (6), 194
[5] J.O. González, M.C. Mochón, and F.B. Rosa de la, *Microchimica Acta*, **2005**, 151(1-2), 39
[6] S.V. Sakariya, R.B. Maridia, S.P. Chauhan, B.N. Suhagia, *Int. J. Pharm. Res. Scholars*, **2015**, 4(2), 355
[7] Z.G. Khan, A.S. Patil, and A.A. Shirkhedkar, *Int. J. Spectrosc.*, **2014,** 2014, 1
[8] ICH Harmonized Tripartite Guideline, Q2 (R1): Validation of Analytical Procedures: Text and Methodology, International Conference on Harmonization, Geneva, Switzerland, **2005**.