Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Der Pharmacia Lettre, 2012, 4 (6):1837-1842
(http://scholarsresearchlibrary.com/archive.html)

Der Pharmacia Lettre
Scholars Research Library
ISSN 0975-5071
USA CODEN: DPLEB4

Estimation of norfloxacin in tablet dosage form by using UV-Vis spectrophotometer

Inamullah[a], Sunil Singh[a]*, Surabhi Sharma[b], Ajit Ku. Yadav[c], Hemendar Gautam[d]

[a]Department of Pharmaceutical Chemistry, Invertis Institute of Pharmacy, Invertis University, Bareilly-243 123, Uttar Pradesh, India
[b]Department of Pharmacognosy, Invertis Institute of Pharmacy, Invertis University, Bareilly-243 123, Uttar Pradesh, India
[c]Department of Pharmaceutics, Invertis Institute of Pharmacy, Invertis University, Bareilly-243 123, Uttar Pradesh, India
[d]Department of Pharmacology, Invertis Institute of Pharmacy, Invertis University, Bareilly-243 123, Uttar Pradesh, India

---

ABSTRACT

A simple, accurate, sensitive and precise Ultraviolet specrophotometric method has been developed for the determination of Norfloxacin in tablet dosage form. The solutions of standard and sample were prepared in 0.1 N Hydrochloric acid. In the UV specrophotometric method, the quantitative determination of the drug was carried at 277 nm and the linearity range was found to be 2-12 µg/ml. For the first order derivative specrophotometric method, the drug was determined at 265 nm with the linearity ranges 2-12 µg/ml. The calibration graphs constructed at their wavelength of determination were found to be linear for UV and derivative specrophotometric methods. The proposed methods have been extensively validated statistically that included parameters such as linearity, accuracy, precision, LOD, LOQ, recovery and robustness. There was no significant difference between the performance of the proposed method regarding the mean values and standard deviations. The described methods can be readily utilized for analysis of pharmaceutical formulation.

Key words: Method development; Validation; Derivative Spectroscopy; Norfloxacin.

INTRODUCTION

Norfloxacin[1], chemically known as 1-ethyl-6-fluoro-4-oxo-7-piperazin-1-yl-1H-quinoline-3-carboxylic acid (**Figure 1**), is a fluorized quinolone, inhibits, like the other members of this group, the gyrase of the bacterial DNA. This effect is held responsible for the bactericidal action of norfloxacin[2]. Follows a selection of sensitive bacili: most enterobacteriaceae (E. coli, klebsiellas, etc.), Pseudomonas aeruginosa, and many pathogenic enteric bacteria (Salmonella, Shigella, etc.), but also Neisseria (especially gonococci). Streptococci are partially resistant whereas anaerobic bacteria are completely resistant[3-5]. It is official in Indian Pharmacopoeia.

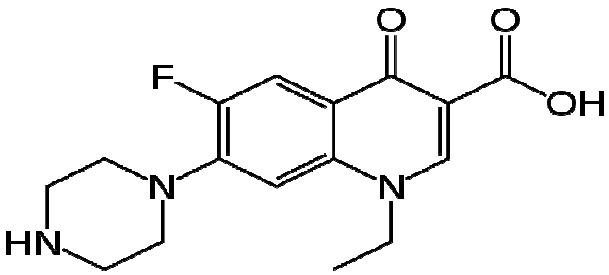

Figure 1- Chemical structure of Norfloxacin

Pharmaceutical research is developing increasingly complex molecules and drug formulations[6-10], and each novel and highly selective analytical technique is therefore of much potential interest. Literature is enriched with several analytical methods for determination of Norflaxacin in single and in other combinations in different solvents[11-17]. A comparison of the results obtained by simple and first order derivative absorption specrophotometric in the ultraviolet region and obtained by HPLC and other instrumental methods of qualitative and quantitative analysis of drugs reveals that simple and first order derivative specrophotometric determinations can be an economically advantageous alternative in many cases[18]. Methods were validated as per the ICH guideline[19]. So, in the present investigation, simple and first order derivative specrophotometric determination of Norfloxacin in tablet dosage form is reported.

MATERIALS AND METHODS

Instrumentation

Analysis carried out on Lab India UV-3200 UV-VIS spectrophotometer, a double beam high speed scanning spectrophotometer (200-800 nm) with a photomultiplier tube detector and having variable spectral bandwidth (0.5-5.0 nm).

Chemicals and reagents

Norfloxacin was received as gratis sample by **Aurobindo Pharma Ltd**, Hyderabad. All chemicals used were of analytical grade (E. Merck, India).

Method. 1-Development of simple spectroscopic method

Standard stock solution

To prepare stock solution of NOR (1000 μg/ml), 100 mg of NOR was placed in 100 ml volumetric flask and dissolved in 75 ml of 0.1 N HCL and the volume was made up to the mark with 0.1 N HCL. 10 ml of the solution was diluted up to 100 ml with 0.1 N HCL to produce final stock solution of 100 μg/ml of NOR.

Sample preparation

Twenty tablets were taken, powdered and powder weight equivalent to 400 mg of NOR was accurately taken and transferred to a 50 ml of volumetric flask. Twenty ml of 0.1 N HCL added to the same and sonicated for 30 min. The flask was shaken, and the volume was diluted to the mark with the same mixture. The above solution was filtered using whatman filter paper no. 1. Appropriate volume of the aliquot was transferred to a 50 ml volumetric flask and the volume was made up to the mark with 0.1 N HCL solution. The spectra were recorded and then measured at 277 nm for NOR. The overlain spectra and calibration curve are shown in **Figure 2 & 3**.

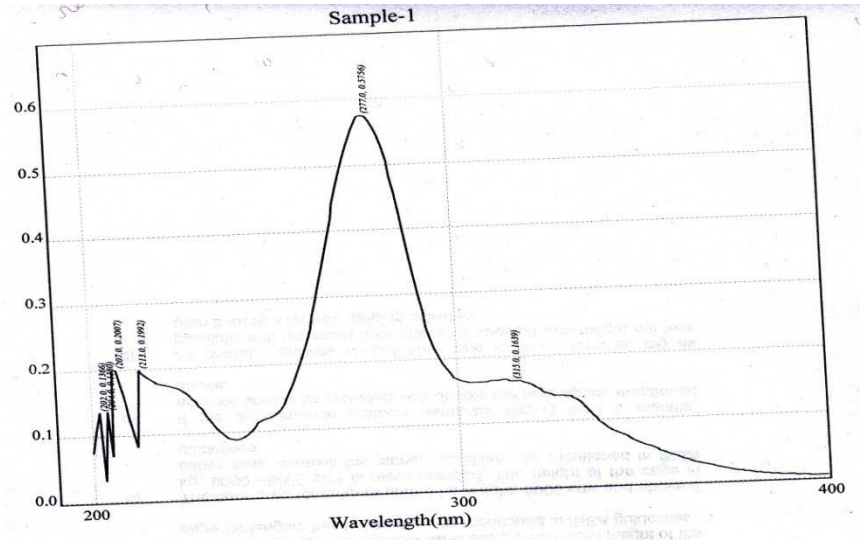

Figure 2- Overlain spectra of Norfloxacin

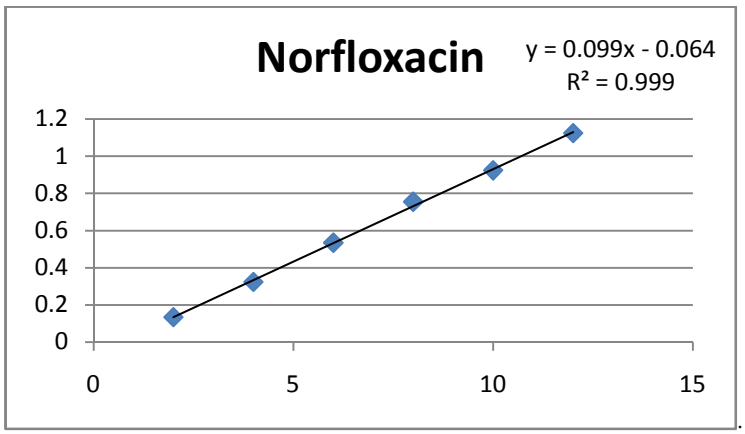

Figure 3- Calibration curve for Norfloxacin

Method. 2-Development of first order derivative method

Standard stock solution

To prepare stock solution of NOR (1000 μg/ml), 100 mg of NOR was placed in 100 ml volumetric flask and dissolved in 75 ml of 0.1 N HCL and the volume was made up to the mark with 0.1 N HCL. 10 ml of the solution was diluted up to 100 ml with 0.1 N HCL to produce final stock solution of 100 μg/ml of NOR.

Sample preparation

Twenty tablets were taken and powdered then powder weight equivalent to 400 mg of norflaxacin was accurately taken and transferred to a 50 ml of volumetric flask. Twenty ml of 0.1 N HCL added to the same and sonicated for 30 min. The flask was shaken, and the volume was diluted to the mark with the same mixture. The above solution was filtered using whatman filter paper no. 1. Appropriate volume of the aliquot was transferred to a 50 ml volumetric flask and the volume was made up to the mark with 0.1 N HCL solution. The first derivative spectra were recorded and then measured at 265 nm for NOR. The overlain spectra and calibration curve are shown in **Figure 4 & 5**.

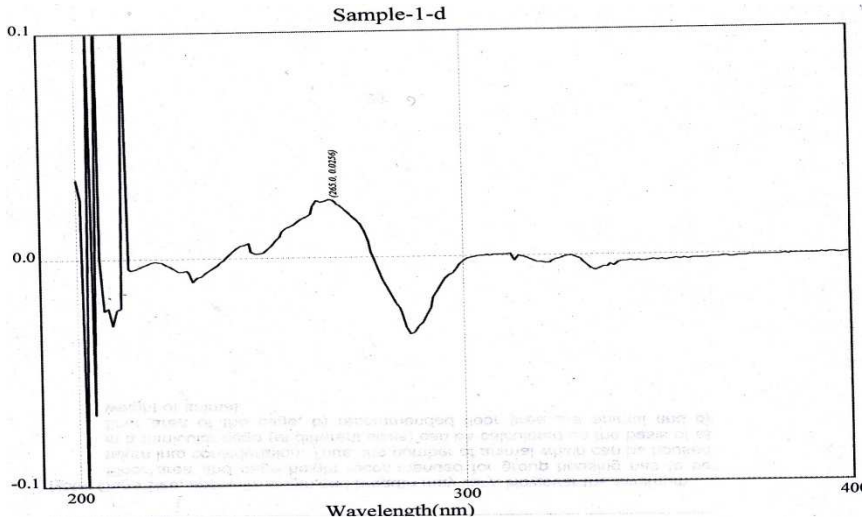

Figure 4- Derivative spectra for Norfloxacin

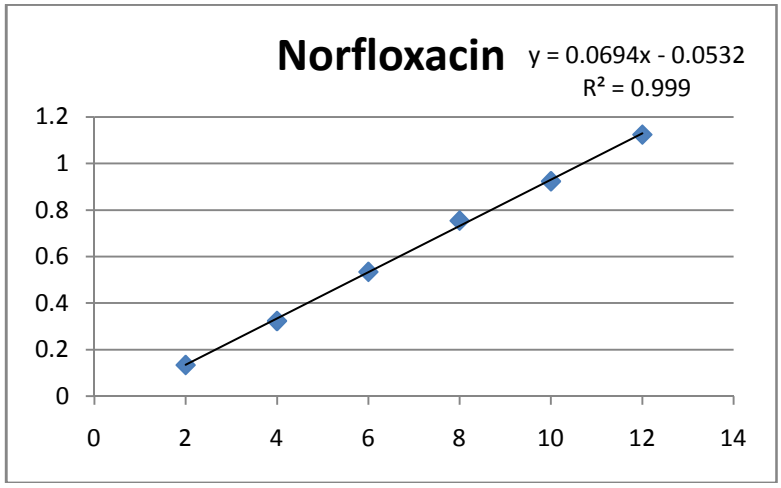

Figure 5- Calibration curve for Norfloxacin

Linearity

Different aliquots were pipette out from standard stock solution into a series of 10 ml volumetric flasks and the volume was made up to the mark with 0.1 N HCL to get concentrations of 2, 4, 6, 8, 10, and 12 μg/ml of norfloxacin. The solutions were scanned on spectrophotometer (Lab India-3200) in the UV range 200-400 nm. The linearity was found 0.999 and range was found 2-12 μg/ml for both methods.

Recovery studies

To the preanalyzed sample solutions (10 μg/ml of Norflaxacin), a known amount of standard stock solution were added at different levels *i.e.* 80, 100 and 120%. The solutions were reanalyzed by proposed method.

Precision

Precision is determined by intra-day and interday precision. Intra-day precision was determined by analyzing the 6, 8 and 10 μg/ml of drug solution for three times in the same day for both proposed methods. Inter-day precision was determined by analyzing the 6, 8 and 10 μg/ml of drug solutions daily for over the period of a week for both proposed methods.

Repeatability

Repeatability was determined by analyzing 10 μg/ml concentration of drug solution for six times.

Limit of Detection and Limit of Quantitation

Several approaches for determining the detection limit and quantitation limit are possible, depending on whether the procedure is a non-instrumental or instrumental.

Ruggedness

Ruggedness of the proposed method is determined by analysis of aliquots from homogenous slot by two analyst using same operational and environmental conditions.

RESULTS AND DISCUSSION

In UV spectroscopic method, the spectra were utilized for developing the equations for analysis. Norfloxacin showed maximum absorbance at 277 nm and 265 nm for simple and derivative spectroscopy respectively. The normal spectra were derivatized into first order derivative, using UV software of instrument, where $\Delta\lambda = 2$. The amplitudes of the corresponding troughs were measured at 265 nm (**Table 1**). The percentage recovery value obtained within standard limit of 99.23% to 101 % for both methods which confirmed that the methods were accurate and free from any interference of excipients (**Table 2**). The low value of standard deviation obtained indicated precision of the method. Results of intraday and interday precision studies are reported in **Table 3**. The reproducibility, repeatability (**Table 4**) and ruggedness (**Table 5**) of proposed methods were found to be satisfactory which was evidenced by low values of standard deviation, LOD and LOQ (**Table 6**) for both methods were found to be satisfactory.

Table:1-Linearity study of Norfloxacin

Sr. No.	Concentration μg/ml	Method 1		Method 2	
		Amplitude (Mean ± SD)	% RSD	Amplitude (Mean ± SD)	% RSD
1	2	0.1869±**0.023**	0.98	0.132±**0.12**	1.02
2	4	0.4674±**0.012**	0.87	0.321±**0.31**	1.13
3	6	0.7121±**0.041**	1.02	0.523±**0.02**	0.97
4	8	0.9946±**0.033**	0.99	0.843±**0.06**	0.99
5	10	1.3131±**0.021**	1.21	1.132±**0.21**	1.03
6	12	1.5359±**0.015**	1.09	1.421±**0.07**	1.09

Table:2- Results of recovery studies

Sr.No.	Pre-analysed sample solution (μg/ml)	Method 1		Method 2	
		% Amount of drug added (μg/ml) (n=3)	% Recovery	% Amount of drug added (μg/ml) (n=3)	% Recovery
1	8	80%	99.21±0.32	80%	101.02±0.48
		100%	98.96±0.41	100%	99.97±0.21
		120%	100.24±0.34	120%	100.02±0.14

Table:3- Results of precision studies (Intra-day and Inter-day)

Component	Conc. μg/ml	Method 1				Method 2			
		Intra-day precision		Inter-day precision		Intra-day precision		Inter-day precision	
		Conc. found (n=3)	RSD	Conc. found (n=3)	RSD	Conc. found (n=3)	RSD	Conc. found (n=3)	RSD
Norfloxacin	6	5.9±0.53	1.02	6.03±0.23	0.95	5.7±0.51	1.21	5.8±0.24	1.21
	8	7.4±0.42	0.98	7.65±0.31	0.89	8.04±0.41	1.04	8.7±0.61	0.98
	10	9.6±0.46	1.05	9.96±0.54	0.93	9.73±0.52	0.99	9.8±0.57	0.95

Table:4- Results of repeatability studies

Component	Method 1			Method 2		
	Amount taken (μg/ml) (n=6)	Amount found (%)	RSD	Amount taken (μg/ml) (n=6)	Amount found (%)	RSD
Norfloxacin	10	99.55 ± 0.87	1.32	10	99.86 ± 0.75	1.47

Table:5- Results of ruggedness studies

Component	Label claim (mg)	Method 1 Amount Found (%) (n=5)		Method 2 Amount Found (%) (n=5)	
		Analyst I	Analyst II	Analyst I	Analyst II
Norfloxacin	400	101.14 ± 0.65	99.33 ± 0.87	98.99 ± 0.64	100.33 ± 0.47

Table:6- Results of LOD and LOQ

Component	Method 1		Method 2	
	LOD	LOQ	LOD	LOQ
Norfloxacin	0.46	1.87	0.32	1.44

REFERENCES

[1] Anonymous *Indian Pharmacopoeia*. **2007**; 3: 841.
[2] Patel K, Singh S, Sahu P, Trivedi P. *Scholars Research Library, Der Pharmacia Lettre*. **2011**; 3 (6):102-107.
[3] Singh S, Yadav AK, Gautam H. *Bull Pharm Res.* **2011**; 1 (3): 10-2.
[4] Singh S, Dubey N, Jain DK. *International Journal of Biomedical and Phaarmaceutical Sciences.* **2011**; 5 (1): 57-60.
[5] Singh S, Yadav AK, Gautam H. *Bull Pharm Res.* **2012**; 2 (2): 83-6.
[6] Singh S. *Scholars Research Library, Der Pharmacia Lettre*. **2012**; 4 (2): 509-514.
[7] Singh S, Patel K, Agrawal VK, Chaturvedi S. *Scholars Research Library, Der Pharmacia Lettre*. **2012**; 4 (3): 897-905.
[8] Singh S, Dubey N, Jain DK. *Asian J. Research Chem*. **2010**; 3 (4): 885-887.
[9] Borrego CM, Diaz CM. *J Pharma and Biomed Anal.* **1999**; 18: 919-926.
[10] Ghante RM, Pannu KH, Loni A. *IJPPS*. **2012**; 4 (4): 241-245.
[11] Rege VP, Sathe AP. *Int J Adv in Pharma Res.* **2011**; 2 (11): 592-597.
[12] Sebaiy MM, Abdullah A. *Asian J Pharma*. **2011**; 1 (4): 79-84.
[13] Patel P, Patel K, Bhatt K. *Int J Res in Pharma and Biomed Scien*. **2011**;.2 (2): 710-713
[14] Abou-Taleb HN, Ei-sherbiny. *Int J Biomed Scie*. **2011**; 7 (2): 137-144.
[15] Bedor GCD, Goncalves MT. Braz J Pharma Scie. **2007**; 43 (2): 231-238.
[16] Galaon T, Udrescu S. *Chromatographya*. **2007**; 21(1): 40-47
[17] Lim J, Park B. J Chromatogr B. **2002**; 7 (1): 185-189.
[18] Samanidou VF, Christodoulou EA. *Bioanal Chem*. **2003**; 375 (2): 623-629.
[19] ICH, Stability Testing of New Drug Substances and Products Q1A (R2), *International Conference on Harmonization*, IFPMA, Geneva, **2003**.