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ANALYTICAL METHOD DEVELOPMENT AND VALIDATION OF NORFLOXACIN IN BULK AND DOSAGE FORMS USING UV SPECTROPHOTOMETRY

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ABSTRACT

An attempt was made to develop simple and economical methods for the estimation of norfloxacin by UV method. In absorption 278 nm was selected wave length. The statistical analysis of data obtained for the calibration curve of norfloxacin in pure solution indicated a high level of precision for the proposed method, as evident by low relative standard deviation. The correlation coefficient was found to be significant. The linearity range showed straight line passing through origin. The method was validated by accuracy, precision and low values of % RSD results of recovery studies also proves the accuracy of method. For UV method methanol is selected as solvent which shows a maximum absorbance than other solvents like sodium hydroxide, HCL. The linearity was obtained for norfloxacin 4-16gm/ml. The precision was confirmed by low values of S.D and % RSD less than 2% conclude that no changes in validation parameter.

KEYWORDS: Norfloxacin, UV method, Linearity**,** recovery.

INTRODUCTION

Pharmaceutical analysis in general terms comprises those procedures necessary to determine the identity, strength, quality, purity of drugs and chemicals. Particularly, it becomes necessary to broaden the scope of definition to include analysis of raw materials, intermediate in manufacturing of drugs biological samples, etc. The analytical chemistry based on sound scientific principles involves suitable combination of chemistry and mathematics. This has led to tremendous increase in the responsibilities and challenges of a pharmaceutical analyst. Analytical chemistry is defined as the science and art of determining the composition of materials in terms of elements of compounds which they contain. It refers to all techniques and methods for identifying constituents in sample Analytical chemistry is divided into two types as follows.

1. Qualitative Analysis: It is concerned with the nature and the kind of material in the sample, without specific interest in the exact amount present.

2. Qualitative Analysis: It is concerned with the amounts of various materials in the sample and the results of such analysis are expressible in terms of numbers.

The study of analytical chemistry provides ideal training for nearly all scientists course in quantitative analysis equips the analyst with the ability to record and intercept such word, and understood to communicate results. These methods are generally quite rapid where hundreds of samples are analysed as a routine. The main drawback of high instrument cost can be compensated by operational cost and increased output. Among the instrumental analytical techniques, the UV visible spectrophotometer methods are comparatively, simpler, sensitive, rapid and may be adapted for the analysis of binary mixtures without prior separation.

Drug profile

Norfloxacin is a synthetic antibacterial agent that belongs to the class of fluoroquinolone antibiotics. It is used to treat urinary tract infections, gynecological infections, inflammation of the prostate gland, gonorrhea and bladder infection.

Norfloxacin is a synthetic antibacterial agent that belongs to the class of Fluoroquinolone antibiotics. It is used to treat urinary tract infections, Gynecological infections, inflammation of the prostate gland, Gonorrhea and bladder infection. Eye drops were approved for use in children older than one year of age. It is sold under the brand name Noroxin.

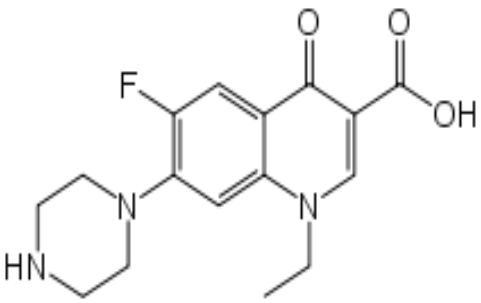

Fig. 1: Structure of Norfloxacin.

MATERIALS AND INSTRUMENTS

All chemicals are analytical grade.
Norfloxacin was obtained as a gift sample from HETERO DRUGS, Hyderabad and formulation brand name NORFLOX (containing of norfloxacin) it was purchased from local pharmacy.

Instrument: Uv- Visible Spectrophotometer (Uv
Experimental work
Selection of solvent
As per solubility and observation of spectrum of the following solvents were selected for the trail basis, as shown in Table 6.2.1.

Preparation of standard drug solution
Accurately weighed 10mg of NORFLOXACIN was transferred into 100ml of volumetric flask. It was dissolved upto the mark with 0.1N HCL to obtain stock solution (100µg/ml).

The solution in concentration ranging from 0.2 µg/ml to 1µg/ml was taken in 10ml volumetric flask. It was dissolved upto the mark with 0.1N HCL they are prepared to mark dilutions. The determination was conducted 6 times at room temperature.

Fixation of suitable parameters for the drug
Location of λ max
The standard stock solution of NORFLOXACIN was appropriately diluted with 0.1N HCL to get the concentration of 0.2µg/ml to 1µg/ml respectively. The solutions were scanned in the range (400nm to 200nm) against solvent blank. The adsorbance spectra of drug is depicted in fig 1 norfloxacin has shown maximum absorbance at 278nm & it was selected for method.

Determination of Norfloxacin adsorption at selected wavelength
Quantitative estimation of norfloxacin absorption was determined at the selected wavelength (278nm).

Study of beers lamberts law
The standard stock solution of norfloxacin was diluted with 0.1N HCL to get series of standard concentration from (0.2µg - 1µg). absorbance of each of these solutions were measured at 278nm using blank. The graph plotted as concentration Vs absorbence is depicted in Fig 6.2.5.

Analysis of tablet formulation by proposed methods
10 tablets were weighed to obtain the average tablet weight, which were then powdered .powder equivalent to 100 mg of NORFLOXACIN was weighed & transferred to 50ml volumetric flask & allowed to dissolve in 50ml of 0.1N HCL. The volume was made upto the mark with 0.1N HCL to get the solution having norfloxacin 100µg/ml & the sample solutions were prepared with in the concentration range by using 0.1N HCL .The results shown in the Table 6.2.6.

Validation of the proposed method
Accuracy
Use a minimum of three spiking concentration in the excipient solution. Prepare 2 samples of each concentration. Test 6 samples in the triplicate on one run. Measure expected Vs average measured value calculate the % recovery = bias.

The total amount of drug was calculate by correcting the absorbance at 278 nm. The percent recovery was then calculate by using following formulae.
% Recovery = T-C / P x 100

Where,
T= total drug estimated
C= Amount from pre analysed tablet powder
P= Amount of pure drug added.

Precision (% RSD)
Prepare three dilutions of the sample (high /medium/low concentration in the range).

Test triplicates of each dilution of the sample in the three different assay.
Do for day-to-day variations
Do for lot-to-lot variations of assays materials
Do for technician-to-technician variations
Calculate the average and standard deviation for each point on the curve for each individual test calculate the %RSD for each point on the curve between the assay runs.

Standard deviation
s=(√(Σ(xi-x^')) 2)/(N-2)
Where,
S=standard deviation
X=observe values
X'=arithmetic mean =ΣX/N
N= Number of deviations
%RSD=s/x×100

Linearity
Determining the co efficient of correlation R for dilutions of sample the range claimed for the assay.

Prepare 6 to 8 samples dilutions across the claimed range.

Test each dilution in triplicate for 3 runs.

Record expected values, actual values and % recoveries for each run.

Analyze each set of dilutions as a linear curve and calculate correlation coefficient (t) for each assay.

r=(Σ(x-x^2)(y-y^2))/√(Σ(x-x^2)2sΣ(y-y^2)^2)

Limit of Detection (LOD)
It is lowest amount of the analyte in a sample that can be detected but not necessary be quantitated as exact concentration or amount,
LOD= 3.3×standard deviation of regression line÷slope

Limit of Quantitation (LOQ)
It is lowest amount of the analyte in a sample that can be measured quantitatively in a sample with acceptable accuracy and precision.

LOQ = 10×standard deviation of low concentration ÷slope

RESULTS AND DISSCUSSION

The solubility of Norfloxacin was determined in a variety of solvents, sample 10mg was taken in a test tube and various solvents were added to check the solubility. The solvents used are DMSO, NaOH, HCL, methanol, ethanol, chloroform. Solubility of Norfloxacin is given in a table 7.2.1.

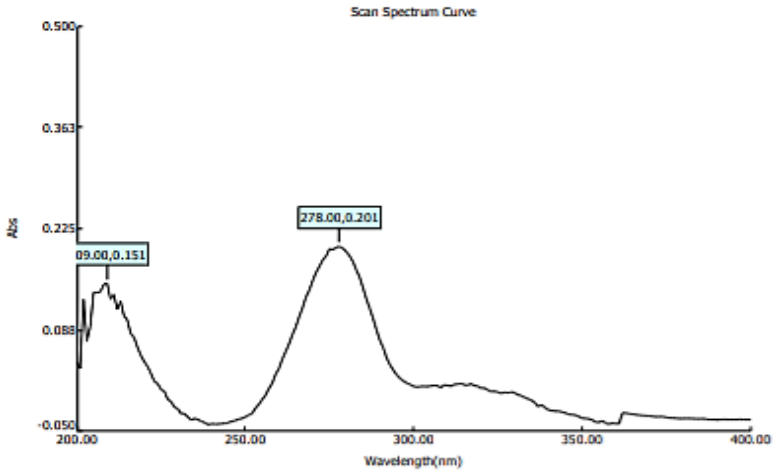

Figure 2: UV Spectrum for norfloxacin.

Based on solubility studies 0.1N HCL and DMSO were chosen as solvent. Based upon its availability and the stability condition in account we selected solvent.

Drug was dissolved in 0.1N HCL and was made further dilutions to produce 10µg/ml. It was scanned in the range of 200nm- 400mn and it show constant λ max at 278 nm this is shown in fig..... stability of the absorbance is checked at their λ. The linearity of the drug norfloxacin was found, it's calibration curve was constructed and shown in figure The optical characteristics such as Beer's law limit (1µg/ml), correlation coefficient (0.9841), slope (0.9702x) were calculated and shown in table 7.2.3.

The limit of detection and limit of quantification were determined from the linearity studies. The limit of detection was found to be 1 µg/ml and the limit of quantification was found to be µg/ml. It has be shown in table 7.2.4.

Linearity and Range
The study was performed over the series of concentration ranging from 0.2 - 1µg/ml for Norfloxacin. The graphs of concentration vs absorbance found to be straight line.

Concentration range (µg/ml)	Absorbance at 278 nm
0	0
0.2	0.201
0.4	0.298
0.6	0.561
0.8	0.768
1	1.024

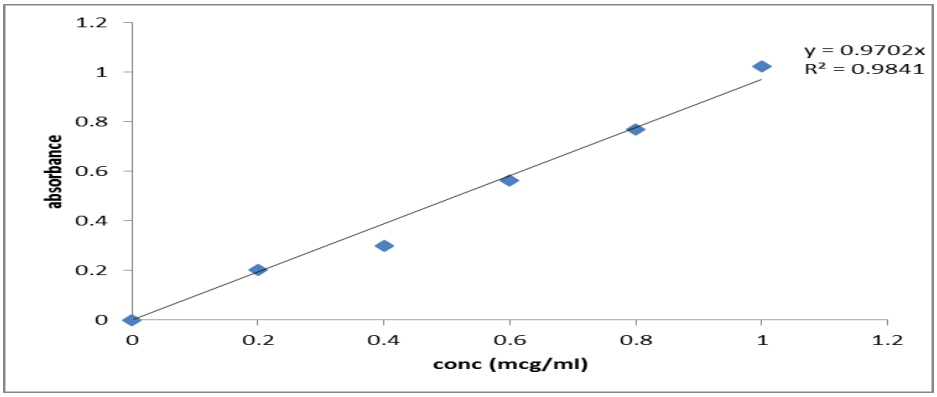

Figure 2: Calibration curve of Norfloxacin.

Accuracy

Accuracy of developed method was determined by recovery study in 3 concentration levels by replicate analysis n=3 Standard drug solutions were added to pre analyzed sample solution and percentage of total drug content was calculated. The analyzed values are shown in tabulated.

Drug sample	Concentration of standard drug	Amount recovered	Percent drug recovery
10 µg/ml	2µg/ml	11.9	99.1
10 µg/ml	4µg/ml	13.8	99.2
10 µg/ml	6µg/ml	16.2	101.2

Precision

Precision of analytical method is expressed as SD and %RSD of any measurements. Precision of estimation of norfloxacin by proposed method was ascertained by replicate analysis of drug norfloxacin. The results are shown in table 7.3.

Limit of detection (lod) and limit of quantification (loq)

LOD AND LOQ can be determined by the method as per ICH guide lines. The method is used in this project is based on standard deviation of response and the slope of calibration curve. The values are shown in table 7.4.

Assay

Formulation	Sample no	Label claim (mg)	Amount obtained	Assay value	%RSD
Noralox	1	400	398.89	99.73	39.78%
	2	400	396.51	99.13	
	3	400	400.10	100.03	

SUMMARY

An attempt was made to develop simple and economical methods for the estimation of norfloxacin by UV method. In absorption 278 nm was selected wave length. The statistical analysis of data obtained for the calibration curve of norfloxacin in pure solution indicated a high level of precision for the proposed method, as evident by low relative standard deviation. The correlation coefficient was found to be significant. The linearity range showed straight line passing through origin. The method was validated by accuracy, precision and low values of % RSD results of recovery studies also proves the accuracy of method. For UV method methanol is selected as solvent which shows a maximum absorbance than other solvents like sodium hydroxide, HCL. The linearity was obtained for norfloxacin 4-16gm/ml. The precision was confirmed by low values of S.D and % RSD less than 2% conclude that no changes in validation parameter.

CONCLUSION

The developed UV-spectroscopic derivative spectroscopic method gives a sensitive, accurate, precise and economical, results for determination of norfloxacin in bulk drug and marketed formulation and easily applied for routine analysis. the economical analytical method was developed for norfloxacin. the most striking features of these methods is it's simplicity and rapidity. The developed methods were successfully applied for determination of the drug in commercial formulation.

REFERENCES

1. Anonymous www.analyticalchem.html.
2. Skoog. DA and West, DM. Fundamentals of analytical chemistry, 8th edition. Thompson Asia Pvt Ltd., Singapore, 2005; 314.
3. Mendham, Vogel's text book of quantitative chemical analysis, 5th edition. British cataloguing in publication data, 1989; 10.

4. Sharma, Y.R. Elementary organic spectroscopy. 4th revised and enlarged edition, S.Chand and company ltd., New Delhi, 2007; 11.
5. Silverstein, R.M., Claytion Bassler. G. and Terrence C. Morrill. Spectrometric identification of organic compound. Lohn Wiley and sons, New York, 1991; 289.
6. Jag mohan. Organic analytical chemistry theory and practice. 2nd edition, Narosa publishing house, New Delhi, 2006; 125-126.
7. Soujanya, BK instrumental method of chemical analysis. 17th edition., Krishna Prakashan media Pvt Ltd., Meerut, 1997; 8.
8. Beckrett, A.H. and stenlake, J.B. practical pharmaceutical chemistry, part B. 4th edition., CBS publishers and distributors, New Delhi, 2002; 272-280.
9. Sethi, P.D. HPLC quantitative analysis of pharmaceuticals formulations. 1st edition, CBS Publishers and distributors, New Delhi, 2001; 3-94.
10. Anonymous www.enwikipedia.com
11. Chatwal, R. Gurudeep and Sham, K.Anand. instrumental methods of chemical Analysis. 5th revised edition., Himalaya publishing house, Mumbai, 2002; 2: 160.
12. Devala rao, G. Text book of pharmaceutical analysis. 4th edition., birla publications pvt. Ltd., 2007-2008; 2: 1-15.
13. Code Q2A, Text on validation of analytical procedures, international conference on hormonization, geneva, October, 1991; 1-5.
14. Code Q2B, Validation of analytical procedures, methodology international conference on hormonization, geneva, November, 1996; 1-8.
15. Sarojkanta Bisoyi*, SudhirKumar Sahoo, Amar Kanta Goudoo. Development And Validation Of A Uv-Vid Spectrophotometric Method For Determination Of Norfloxacin In Bulk And Its Dosage Form And Stability Studies.
16. Inamullaha, Sunil Singha*. V.et .al.., Estimation Of Norfloxacin In Tablet Dosage Form By Using Uv-Vis Spectrophotometer, 2012.
17. Chierentin*and H. R. N. Salgado v.et.al.., Performance Characteristics Of Uv And Visible Spectrophotometry Methods For Quantitative Determination Of Norfloxacin In Tablets, 2014.
18. Jelamvazir*, Minal Rohit and Dr. Dhananjay Meshramv. et.al, 2015.
19. Khushboo Nayak*, Navin K. Khare, AtulSayare, Prashant Ghode, Raymond M. Lawrence v.et.al.., New Validated Uv Spectrophotometric Methods For Estimation Of Norfloxacin And Tinidazole In Bulk And Tablet Dosage Forms, 2012.
20. Sharma Ravi*, Kaur Amandeep v.et.al Simultaneous Estimation Of Loperamide Hydrochloride And Norfloxacin By Validated Uvspectrophotometric Method, 2014.
21. Mayank Pant*, Khemchand Dadare and N. C. khatri application of uv spectrophotometric methods for simultaneous estimation of norfloxacin and tinidazole in bulk and tablet dosage forms.
22. S. B. wankhede*, A. prakash, B. kumari and S. S. chitlange V.et.al Simultaneous Spectrophotometric Estimation Of Norfloxacin And Ornidazole In Tablet Dosage Form, 2009.
23. S. Harshini, M.Aktiful Haque, Dibyalochan mahanty, V.Roja kumari, M. Pravallika, S. Prasanna kumari v.et.al Simultaneous Estimation Of Norfloxacin And Metronidazole In Bulk And Pharmaceutical Dosage Form |By Using Uv Spectrophotometric Method, 2014.