International Journal of Pharmaceutical Research and Applications
Volume 9, Issue 1 Jan-Feb 2024, pp: 1027-1032 www.ijprajournal.com ISSN: 2249-7781

Analytical Method Development and Validation for Estimation of Rantidine Hcl in Solid Dosage Form By Uv-Spectrophotometric Method

CH.Sri Lakshmi[1], D Rama Brahma Reddy[2], M.Lakshminarayana[3], P.Naga Raju[4], P.Vamsi Krishna[5], R.Chandra shekar[6], S.P.Manohar[7.]

[1]*Associate professor Nalanda Institute Of Pharmaceutical Sciences,Siddharth nagar, kantepudi (V),Sattenapalli (M),Guntur (Dist)-522438.*

[2]*Principal&professor Nalanda Institute Of Pharmaceutical Sciences,Siddharth nagar, kantepudi (V),Sattenapalli (M),Guntur (Dist)-522438.*

Students of Nalanda Institute Of Pharmaceutical Sciences,Siddharth nagar, kantepudi (V),Sattenapalli (M),Guntur (Dist)-522438.

Submitted: 25-01-2024 Accepted: 03-02-2024

ABSTRACT

A rapid, simple, selective and precise UV- Visible Spectrophotometric method has been developed for the determination of Ranitidine Hydrochloride in bulk forms and tablet dosage formulations. The spectrophotometric detection was as per carried out at an absorption maximum of 322 nm using 0.1N HCl as solvent. The method was validated for specificity, linearity, accuracy, precision, robustness and ruggedness. The detector response for was linear over the selected concentration range 2-14 ug/ml with a correlation coefficient of 0.9984. The accuracy was carried out as per recovery study and found between 98% to 102%. The results demonstrated that the excipients in the tablets did not interfere with the method and can be conveniently employed for routine quality control analysis of Ranitidine Hydrochloride in bulk and in dosage formulations.(1)

KEYWORDS:UV Spectroscopy; Method Development; Validation; Ranitidine Hydrochloride, 0.1N HCl and ICH Guideline.(2)

I. INTRODUCTION

A spectroscopy method is the branch of science dealing with the study of interaction between Electromagnetic radiation and matter. It is the most powerful tool available for the study of atomic and molecular structure/s and is used in the analysis of a wide range of samples.Spectrophotometric technique is simple, rapid, moderately specific and applicable to small quantities of compounds. The fundamental law that governs the quantitative spectrophotometric analysis is the Beer- Lambert law.

Lambert's law: It states that the intensity of a beam of parallel monochromatic radiation decreases exponentially as it passes through a medium of homogeneous thickness. A combination of these two laws yields the Beer-Lambert law

The validation of an analytic method demonstrates the scientific soundness of the measurement or characterization. It is required to varying extents throughout the regulatory submission process. The validation practice demonstrates that an analytic method measures the correct substance, in the correct amount and in the appropriate range for the samples. It allows the analyst to understand the behavior of the method and to establish the performance limits of the method

Accuracy

It is the closeness of agreement between the values found. The value accepted as a conventional true value or the accepted reference value. Several methods of determining accuracy are available. It can be screened by the use of an analytical procedure to an analyte of known purity, by comparison of the results of purity, by comparison of the results of the proposed analytical procedure with those of a second accepted procedure, the accuracy of which is stated and defined. It can also be inferred once precision, linearity and specificity have been established.

Precision

Precision of an analytical procedure expresses the closeness of agreement between a series of measurements obtained from multiple sampling of the same homogeneous sample under

International Journal of Pharmaceutical Research and Applications
Volume 9, Issue 1 Jan-Feb 2024, pp: 1027-1032 www.ijprajournal.com ISSN: 2249-7781

the prescribed conditions. It can be sub divided into repeatability, intermediate precision and reproducibility.

- **Repeatability** should be assessed using a minimum of 9 determinations covering the specified range for the procedure by 3 replicates or 6 determinations at 100% of the test concentration.
- **Intermediate precision** depends upon the circumstances under which the procedure is intended to be used. The specific day, analyst performing, equipment are the random events that cast effect on the precision of the analytical procedure. It is not considered necessary to study these effects individually. The use of an experimental design should be encouraged. Reproducibility is assessed by means of an inter-laboratory trial.
- **Reproducibility** should be considered in case of the standardization of an analytical procedure.

Specificity

Is the ability to assess the analyte for the presence of various components that may be present. It can be established by a number of approaches, depending on the intended purpose of the method. The ability of the method to assess the analyte of interest in a drug product is determined by a check for interference by placebo. Specificity can be assessed by measurement of the API in samples that are spiked with impurities or degradants. The detection limit of an individual analytical procedure is the lowest amount of analyte in a sample that can be detected. It can be determined visually by signal to noise ratio, standard deviation of the response and the slope.

Detection limit

Signal to noise approach can only be applied to analytical procedures which exhibit baseline noise. Comparing measured signals from samples with known concentrations of analyte with those of blank samples and establishing the minimum concentration at which the analyte can be reliably detected. A signal to- noise ratio between 3 or 2:1 is generally considered acceptable for estimating the detection limit. The detection limit (DL) may be expressed as: DL=3.3 σ/ S where, σ is the standard deviation of the response, S is the slope of the calibration curve. Slope S may be estimated from the calibration curve of the analyte. The estimate of σ may be carried out in a variety of ways, based on the standard deviation of the blank and the calibration curve.

The linearity

An analytical procedure is its ability to obtain test results that are directly proportional to the concentration of analyte in the sample. Test results should be evaluated by appropriate statistical methods, by calculation of a regression line like by the method of least squares. correlation coefficient, y-intercept, slope of the regression line and residual sum of squares for which a minimum of five concentrations are recommended

The range

An analytical procedure is the interval between the upper and lower concentration of analyte in the sample for which it has been demonstrated that the analytical procedure has a suitable level of precision, accuracy and linearity. The ICH guidance on validation distinguishes the types of methods according to the purpose of the method and lists suitable evaluation type. The ICH guidelines suggest detailed validation schemes relative to the purpose of the methods. It lists recommended data to report for each validation parameter. Acceptance criteria for validation must be based on the previous performances of the method, the product specifications and the phase of development. (3)

II. MATERIALS AND METHODS

Instruments

The analysis was performed by using the analytical balance (Mettler), pH meter (Cyber scan), UV spectrophotometer (UV-Lambda 25, Perkin Elmer equipped with variable wavelength detector and data integration software).

Selection of solvent: 0.1N HCl

Preparation of solvent system

Accurately weighed 100 milligrams of Ranitidine hydrochloride was transferred to previously dried 100 ml volumetric flask and dissolved in 0.1N HCl. The final volume of this stock solution was made up to 100 ml with 0.1N HCl (Solution A). From the stock solution (Solution A) solution was taken (10 ml) and further diluted to 100 ml with 0.1N HCl to make 100 µg/ml concentration (Solution B).

Determination of λ max

To determine the λ max, 100 milligrams of Ranitidine hydrochloride was accurately weighed and transferred to previously dried 100 ml

International Journal of Pharmaceutical Research and Applications
Volume 9, Issue 1 Jan-Feb 2024, pp: 1027-1032 www.ijprajournal.com ISSN: 2249-7781

volumetric flask and dissolved in 0.1N HCl. The final volume of this stock solution was made up to 100 ml with 0.1N HCl (Solution A). From the stock solution (Solution A) solution was taken (10 ml) and further diluted to 100 ml with 0.1N HCl to make 100 µg/ml concentration (Solution B). From this solution (Solution B) the solutions of concentrations 2, 4, 6, 8, 10, 12and 14 µg/ml were prepared. All the solutions were scanned in the range of 200nm to 400nm (Figure 1) and λ max was determined.

Preparation of Standard Curve of Ranitidine Hydrochloride

A standard curve was prepared by using stock solution B (100 µg/ ml). Absorbance were measured for concentrations 2, 4, 6, 8, 10, 12 and 14 µg/ml solutions on UV visible spectrophotometer at 322 nm.

Spectrometry

The solution of pure drug, polymers and optimized formulations were prepared in 0.1N HCl & were filtered through Whatman filter paper No.42. The solutions were scanned in the range between 200nm to 400nm.

Method Validation

The developed method was validated as per ICH guidelines. The parameter ICH assessed were specificity, linearity, range, accuracy, precision (repeatability), LOD and LOQ.

- Linearity Aliquots from Six different concentration of the stock solution of Ranitidine Hydrochloride (1 to 5 ml of 100µg/ml) were transferred into 10 ml standard flasks and made volume using 0.1N HCl. The absorbance of the solutions of different concentrations was measured at 322 nm against 0.1N HCl as blank. Linearity was observed between 2-14µg/ml. Range The range of analytical method was decided for Ranitidine Hydrochloride (40-115µg/ml) was observed between 2-14µg/ml.
- Accuracy To ensure the accuracy known the amount of pure drug were added to solvent and these samples are reanalyze by the proposed method and % recovery was studied It was carried out by adding known amounts of analyte corresponding to the concentration levels and results were expressed as % recovery.
- Precision To evaluate repeatability of the method, pure drug of solution within working limit was analyses and being six times. Precision of method was also demonstrated by intraday and inter day variation studies. Intraday studies repeated requirement of standard and sample solution are made in day and % RSD were calculated. Inter day studies are repeated measurement of standard and sample solution were made on 3 consecutive days and % RSD were calculated. The RSD % is not less than 2.0 and indicated high precision for the proposed method.
- Limit of Detector (LOD) LOD is the lowest amount of analyte in sample that can be easily but not necessarily quantified. It was calculated by using the following formula, DL =3.3α/S Where α : standard deviation. S: slop of calibration curve
- Limit of Quantification (LOQ) LOQ is the lowest amount of analyte in sample that can be easily detected and quantified with suitable precision and accuracy. It was calculated by using following formula, QL =10 α/S Where α: standard of deviation. S: slope of calibration curve. (4)

III. RESULTS AND DISCUSSION

The methods discuss in the present work provide convenient, precise and accurate way for estimation of Ranitidine hydrochloride.in bulk and pharmaceutical dosage form using 0.1N HCl.

Selection of solvent

Solubility of Ranitidine hydrochloride was performed in solvent 0.1N HCl. and UV spectra of drug in 0.1N HCl was recorded. The absorbance value of drug was higher at λmax with 0.1N HCl as a solvent. Hence, 0.1N HCl was selected as a solvent for further investigation as it is more economical. (5)

Determination of Wavelength

An UV Spectrophotometric scanning (200-400) was carried out to select the wavelength (λmax) for detection of Ranitidine hydrochloride. The wavelength of maximum absorbance (λ max) was found to be 322 nm in HCl. .

International Journal of Pharmaceutical Research and Applications
Volume 9, Issue 1 Jan-Feb 2024, pp: 1027-1032 www.ijprajournal.com ISSN: 2249-7781

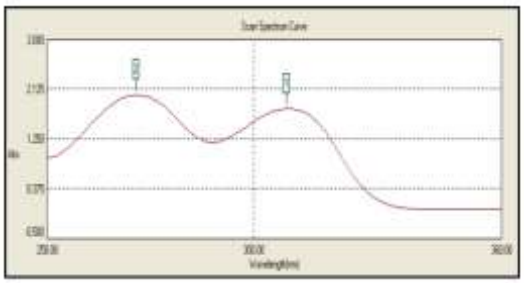

Figure 1 : UV absorption spectrum of Ranitidine Hydrochloride in 0.1N HCl.

Validation of method as per ICH guidelines

The developed method was validated as per ICH guidelines (ICH Q1B, 1996, ICH Q2 R1, 2005) for following parameters. The precision of an analytical procedure expresses the closeness of agreement (degree of scatter) between a series of measurements obtained from multiple sampling of the same homogeneous sample under the prescribed conditions. (6)

Linearity

Eight different concentration of Ranitidine hydrochloride were prepared and analysed at wavelength 322 nm. UV absorption spectrum of Ranitidine Hydrochloride in 0.1N HCl showed λ max at 322 nm. Absorbance obtained for various concentrations of Ranitidine Hydrochloride in 0.1N HCl were given in Table 6.1.

Preparation of Calibration Curve of Ranitidine Hydrochloride

The graph of absorbance vs. concentration for Ranitidine Hydrochloride was found to be linear in the concentration range of 2 – 14 µg /ml was shown in Figure 6.2.

Table: Linearity Range of Ranitidine Hydrochloride in 0.1N HCl.

Sr. No	Concentration(µg)	Absorbance
01	0	0
02	2	0.156±0.12
03	4	0.284±0.24
04	6	0.437±0.20
05	8	0.568±0.24
06	10	0.683±0.22
07	12	0.805±0.18
08	14	0.947±0.23

n=6

Calibration curve for Ranitidine Hydrochloride in 0.1N HCl

Curve of Ranitidine Hydrochloride was carried out at λmax 322 nm in 0.1N HCl. Regression coefficient of Ranitidine Hydrochloride was found to be R2 0.998.the standard linear equation was found to be y= 0.066x+0. 018. Graph shown in Figure.6.2

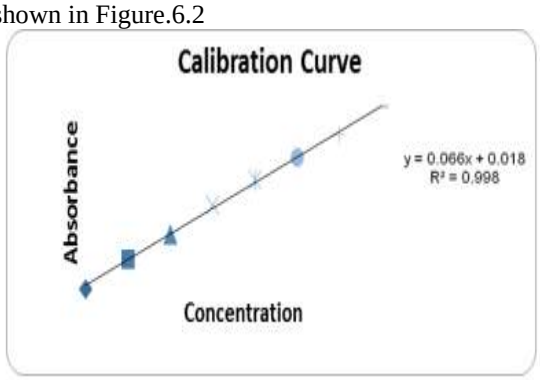

Figure 2 : Standard Curve for Ranitidine Hydrochloride in 0.1N HCl

Optimization parameter of Ranitidine Hydrochloride in 0.1N HCl

The various optimised parameters obtained from the curve of Ranitidine Hydrochloride, carried out at λ max 322 nm in 0.1N HCl are noted in Table 3. 2. Regression coefficient of Ranitidine Hydrochloride was found to be R2 0.998. the standard linear equation was found to be y= 0.066x+0. 018.

Table Optimization parameter of Ranitidine Hydrochloride in 0.1N HCl

Parameter	Method values
Wavelength detection	322 nm
Beers law	2 – 14 µg /ml
Correlation Coefficient	0.998
Regression coefficient	y= 0.066x+0. 018.
Slope	0.066
Intercept	0.018

Accuracy

The concentration 70µg/ml was taken as 80,100,120 and % recovery was found to be range 99%-101%. Hence forward the parameter was found to be validated.

International Journal of Pharmaceutical Research and Applications
Volume 9, Issue 1 Jan-Feb 2024, pp: 1027-1032 www.ijprajournal.com ISSN: 2249-7781

Table : Result of accuracy of Ranitidine Hydrochloride in 0.1N HCl.

Name of Drug	Recovery Level in %	Concentration	Amount Recovered	% Recovery with SD
Ranitidine Hydrochloride	80%	80	79.46	99.32± 0688
	100%	100	100.03	100.03±0.5033
	120%	120	121.08	100.9±0.723

Range

Range is an interval between maximum and lowest concentration limit of the analyte. The drug obeys Beer- Lambert's law in the range of 2 – 14 µg /ml.

Precision

Precision of the method was determined in terms of repeatability and intraday and interday precisions. Repeatability of the method was determined by analysing six samples of same concentrations of drug. Intraday precision was determined by analysing the drugs at three different concentrations and each concentration for three times, on the same day. Interday precision was determined similarly, but the analysis being carried out daily, for three consecutive days. Accuracy, LOD, LOQ and Sandell's sensitivity are determined and the results are summarized in Table. (7)

Table.: Validation in 0.1N HCL.

Sr.no	Parameter	Result	ICH standard	Inference
1	Accuracy	99.54	98%±102%	Pass
2	Precision			
	Interday	➢ 0.8289	RSD<2	Pass
	Intraday	0.00104	-	Pass
3	LOD	0.31380µg/ml	-	-
4	LOQ	0.95093µg/ml	-	-
5	Linearity	0.9998	>0.997	Pass
6	Range	2 - 14 µg /ml.	-	-
7	STD regression	y= 0.066x+0. 018.	-	-

IV. CONCLUSION

- The absorption maximum of Ranitidine Hydrochloride was selected at 322nm for the analysis.
- Regression analysis shows linearity over the concentration range of 2 – 14 µg /ml with correlation coefficient R 2 0.998 (Figure 3.2). The % RSD for repeatability (n=6) precision was found to be less than 2% indicating the precision of method. Accuracy of proposed methods was ascertained by recovery studies and the results are expressed as percentage recovery. Percentage recovery for was found 99.54% which is well within the range between 98 % to 100.2%. The % RSD value for was found to be less than 2%.
- In this study estimation of Ranitidine Hydrochloride was carried out by UV spectroscopy method and all the validation parameters were found satisfactorily. The result of developed method and validation was given in Table.
- The analytical method for estimation of Bisoprolol Fumarate has been developed and validated according to validation protocol of ICH guidelines.
- All parameters mentioned in the protocol were tested and they fulfilled the requirement of ICH analytical method validation for the drug.
- The results obtained are well within the set limit; indicates that the described analytical method is suitable for estimation of Ranitidine Hydrochloride in bulk as well as formulation.

REFERENCES

[1]. R. K. Nanda, S. E. Potawale, V. V. Bhagwat, R. S. Deshmukh, P. B. Deshpande; Development and validation of a HPTLC method for simultaneous Densitometric analysis of Ranitidine hydrochloride and Dicyclomine hydrochloride as the bulk drugs and in the tablet dosage form; 2021, 3(8), 1997-1999.

[2]. Y. X. Chang, Y. Q. Qiu, L. M. Du, C. F. Li, M. Guo; Determination of ranitidine, nizatidine, and cimetidine by a sensitive fluorescent probe; Analyst; 2022, 136(20). 4168-73.

[3]. Vania Maslarska, Determination of ranitidine hydrochloride in pharmaceutical preparations by direct Potentiometriy; 2022, 6(1). 538-540.

[4]. Diane A. I. Ashiru, Rajesh Patel, Abdul W. Basit; Simple and universal HPLC-UV method to determine cimetidine, ranitidine, famotidine and nizatidine in urine: 2020, 860(2), 235–240.

[5]. C. A. Lau-Cam, M. Rahman, R. W. Roos; Rapid Reversed Phase High Performance Liquid Chromatographic Assay Methodcal preparations by ultraviolet and visible spectrophotometry;2023, 26(9). 1933-1941.

[6]. P. Norouzi, M. R. Ganjali, P. A. Daneshgar; novel method for fast determination of ranitidine in its pharmaceutical formulations by fast continuous cyclic voltammetry. 2020, 55(3), 289-96.
[7]. M. H. Bachani, D. S. Acharya, K. V. Shah; Development and validation of HPLC method for simultaneous estimation of dicyclomine hydrochloride, acetaminophen and clidinium bromide in solid dosage form; 2023, 5(2), 462-466.
[8]. R. J. Hedpara, V. Chhatarala, A. Agola; A validated RP- HPLC method for the simultaneous estimation of paracetamol and dicyclomine hydrochloride in pharmaceutical formulations; 2020, 4(6), 2400-2405.
[9]. D. A. Shah, J. P. Rana, U. K. Chhalotiya, S. L. Baldania, K. K. Bhatt; Development and Validation of a Liquid Chromatographic Method for Estimation of Dicyclomine Hydrochloride, Mefenamic Acid and Paracetamol in Tablets; 2021, 76(6), 529–534.
[10]. S. R. Dhaneshwar, A. L. Suryan, V. K. Bhusari, K. S. Rasal; validated HPTLC method for nimesulide and dicyclomine hydrochloride in formulation. 2021, 4(7), 2288-2290..
[11]. Wadher SJ, Kalyankar TM, Panchal PP. Development and validation of simultaneous estimation of chlorpheniramine maleate and phenylephrine hydrochloride in bulk and capsule dosage form by ultra-violet spectrophotometry. 2023:5(5): 2410- 2419.
[12]. Kalyankar TM, Kakade RB, Attar MS, Kamble AR. Simultaneous spectrophotometric estimation of artesunate and mefloquine. Journal of Chemistry. 2020,67,:1-5.
[13]. Kalyankar TM, Wadher SJ, Kulkarni PD. Simultaneous Estimation and Development of UV Spectroscopic Method for Determination of Cinnarizine and Domperidone in Bulk and Pharmaceutical Formulation. 2023: 6 (1):323-329.
[14]. Kalyankar TM, Panchakshari PP, Wadher SJ, Pekamwar SS. Simultaneous Estimation of Duloxetine and Methylcobalamin in combined dosage form by Ultra-violet Spectrophotometry. 2023: 5 (4):1572-1580.
[15]. 21. Kalyankar TM, Kokate RH. Spectrophotometric simultaneous determination of montelukast sodium and desloratidine in combined dosage form by simultaneous estimation method2022: 5 (1):21-24.
[16]. 22. Raj PM, Rao RV, Mukherjee PB, Sarvanan VS, Gopal N, Kalyankar TM. UV-Spectrophotometric Determination of Esomeprazole in tablet dosage forms. 2021:19 (4): 3250-3252.