Available online at: http://www.iajps.com   *Research Article*

DEVELOPMENT AND VALIDATION OF UV SPECTROPHOTOMETRIC ESTIMATION OF CEFADROXIL IN BULK AND TABLET DOSAGE FORM USING AREA UNDER CURVE METHOD

Digge Pradip[1], Jadhav Santosh[2], Gaikwad Sumit[1], Jamagondi Laxmi[1].
1. Department of Pharmaceutics, D.S.T.S College of Pharmacy, Solapur, Maharashtra, India.
2. Department of Pharmaceutics, Sahyadri College of Pharmacy, Methwade, Sangola-413307, Solapur, Maharashtra, India.

Abstract:

A simple, precise, accurate, and economical UV visible spectrophotometric method has been developed for estimation of Cefadroxil drug by AUC method. The standard and sample solutions were prepared by using double distilled water as a solvent. Quantitative determination of the drug was performed at wavelength range 260-266 nm. The linearity was established over the concentration range of 05-25 µg/ml for Cefadroxil with correlation coefficient value of 0.9993. Precision studies showed that % relative standard deviation was within range of acceptable limits. The mean percentage recovery was found to be 99.26%. The proposed method has been validated as per ICH guidelines.

Keywords: Cefadroxil, UV visible spectrophotometry, AUC, Method Validation.

Corresponding Author:

Digge Pradip,
Department of Pharmaceutics,
D.S.T.S College of Pharmacy,
Solapur,
Maharashtra, India.
Email:*diggepradip@gmail.com*

QR code

INTRODUCTION

Cefadroxil (CFD) is chemically, (7*R*)-7-(α-D-4-Hydroxyphenylglycylamino)-3-methyl-3-cephem-4-carboxylic acid [1]. Cefadroxil is a first-generation cephalosporin antibacterial that is para-hydroxy derivative pf cephalexin and is used similarly in the treatment of mild to moderate susceptible infections [2]. Cefadroxil is official in IP, USP and BP. IP [3], USP [4] and BP [5] describe liquid chromatography method for the estimation of cefadroxil. Literature survey revealed several analytical methods chemiluminescence [6], Spectrophotometric [7-10] and HPLC [11-14] have been reported in bulk, pharmaceutical dosage form for determination of Cefadroxil. Hence an attempt has been made to develop new UV-Spectrophotometric (AUC) method for estimation of Cefadroxil in bulk and pharmaceutical formulations with good accuracy simplicity, precision and economy.
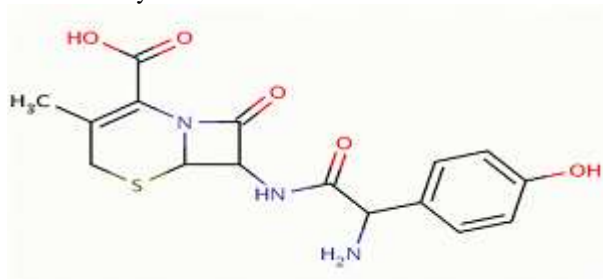

Fig. 1: Chemical structure of Cefadroxil.

MATERIALS AND METHODS

Apparatus and Instrumentation

A shimadzu 1800 UV/VIS double beam spectrophotometer with 1cm matched quartz cells was used for all spectral measurements. Single Pan Electronic balance (CONTECH, CA 223, India) was used for weighing purpose. Sonication of the solutions was carried out using an Ultrasonic Cleaning Bath (Spectra lab UCB 40, India). Calibrated volumetric glassware (Borosil®) was used for the validation study.

Materials

Reference standard of Cefadroxil API was supplied as gift sample by Lupin Laboratory park, Aurangabad. Tablet sample with label claim 250 mg per tablet were purchased from local market Pune.

Method development

Determination of Wavelength Range

For the selection of analytical wavelength range for area under curve method,20µg/ml solution of Cefadroxil was scanned in the spectrum mode from 400 nm to 200nm against distilled water as blank. Wavelength range was selected around wavelength maxima (264nm). Different working standards were prepared between 05-25 µg/ml. Various wavelength range were tried and final wavelength range between 260-266 nm was selected on the basis of linear relationship between area and corresponding concentration (Figure 2).
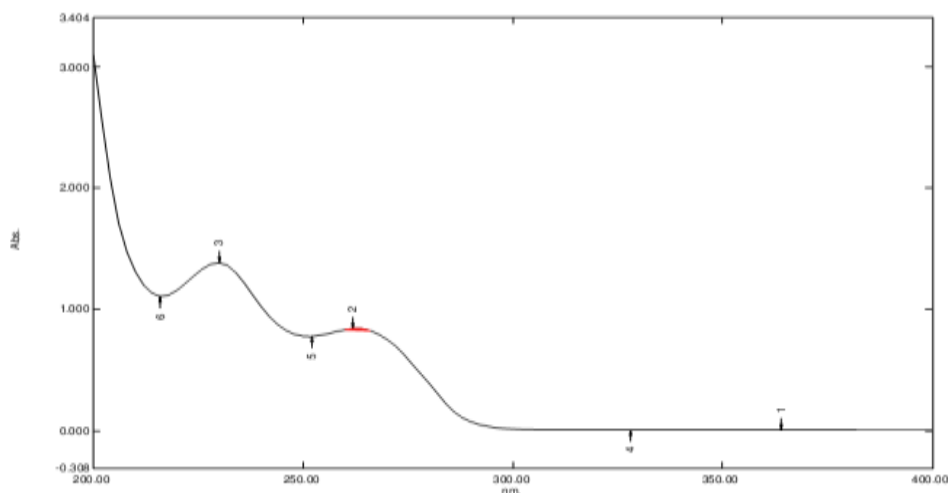

Fig 2: UV AUC Spectrum of Cefadroxil (20µg/ml)

Area under Curve (Area calculation)

Area under curve method involves the calculation of integrated value of absorbance with respect to the wavelength between two selected wavelengths such as λ1 and λ2 representing start and end point of curve region. The area under curve between λ1 and λ2 was calculated using UV probe software. In this study area was integrated between wavelength ranges from 260 to 266 nm.

Area calculation: $(\alpha+\beta) = \int_{\lambda 2}^{\lambda 1} A d\lambda$

Where, α is area of portion bounded by curve data and a straight line connecting the start and end point, β is the area of portion bounded by a straight line connecting the start and end point on curve data and horizontal axis, λ1 and λ2 are wavelength range start and end point of curve region [14].

Preparation of Standard Solution

The standard stock solution of Cefadroxil was prepared by accurately weighing & transferring, 10 mg of API to 100 ml of volumetric flask. The drug was dissolved with sonication in 50 ml of distilled water and volume was made up to the mark by using distilled water. Then take from that 2ml and add to 10ml volumetric flask and make up with distilled water to get final standard stock solution (20µg/ml) was further diluted with distilled water to obtain 05-25 µg/ml Cefadroxil solutions.

Calibration Curve for Cefadroxil

The dilutions were made from Standard Stock solution to get concentration of 05, 10, 15, 20, and 25µg/ml respectively. These solutions were scanned from 400 to 200 nm and area under curve (AUC) values was integrated in the range of 260-266 nm. The calibration curve was plotted between areas under curve values against concentration (Fig. 3).

Assay of Tablet Formulation

Twenty tablets each containing 250 mg of Cefadroxil were weighed crushed to powder and average weight was calculated. Powder equivalent to 10 mg of Cefadroxil was transferred in 100 ml of volumetric flask. A 50 ml of distilled water was added and sonicated for 15 minutes. Then solution was further diluted up to the mark with distilled water. The solution was filtered using Whatmann filter paper no. 41; first 5 ml of filtrate was discarded. This solution was further diluted to obtain 15µg/mL solution with water, subjected for UV analysis using distilled water as blank. This procedure was repeated three times shown in Table 1.
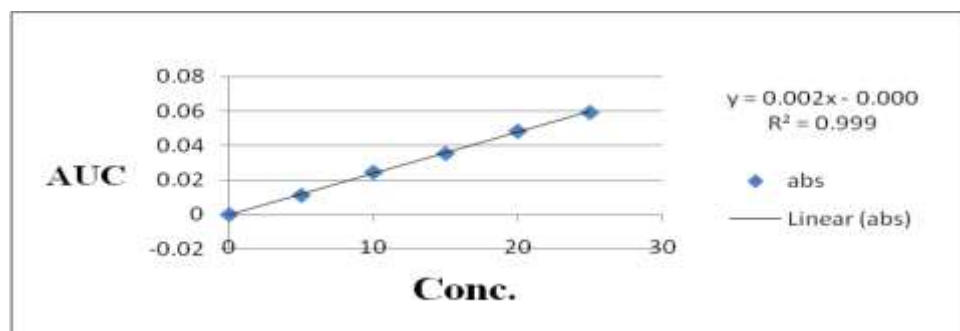

$$y = 0.002x - 0.000$$
$$R^2 = 0.999$$

Fig 3: Linearity of Cefadroxil

Table 1: Assay of tablet dosage form

Sr.No.	Sample Solution Concentration (µg/ml)	Amount found (%)n=3	Mean % found	%RSD
1	15	97.27		
2	15	98.41	97.98	0.6299
3	15	98.25		

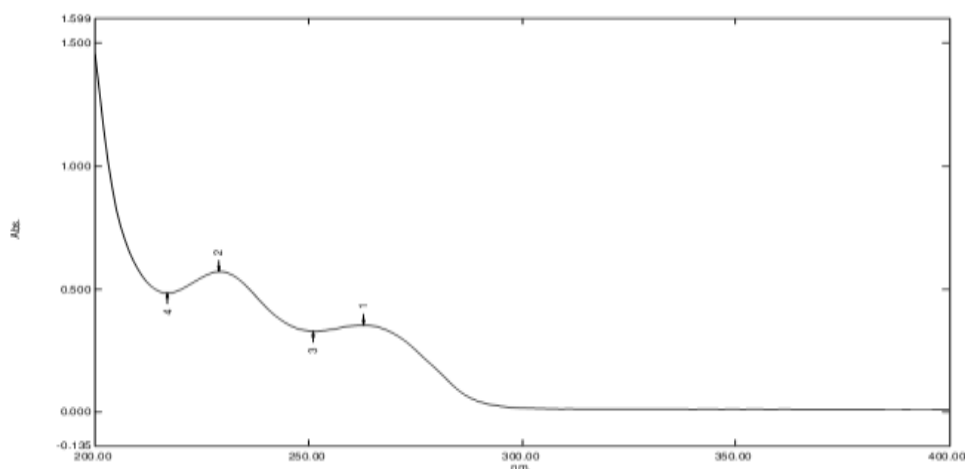*n=3, % RSD = % Relative Standard Deviation.

Fig.4 UV AUC spectrum of Cefadroxil dosage form (15µg/ml)

Method Validation

The above method was validated for various parameters such as Accuracy, Linearity, Precision, Limit of detection (LOD) and Limit of Quantitation (LOQ) according to ICH guideline.

Accuracy

The accuracy for the analytical method was evaluated at 80%, 100% and 120% levels of 20µg/ml standard solution. Area under curve (AUC) was measured in wavelength range 226-240 nm and results were obtained in terms of percent recovery. Three determinations at each level were performed and % RSD was calculated for each level results were shown in table 2.

Precision

The precision of an analytical procedure expresses the closeness of an agreement (degree of scatter) between a series of measurements obtained from multiple sampling of the same homogeneous sample under the prescribed conditions intraday precision was studied by integrating area of standard solution of 20µg/ml concentration at six independent series in the same day. Inter-day precision studies were performed by integrating area of standard solution of 20µg/mlconcentration on three consequent days. The %RSD Was calculated and results were shown in table 3.

Table 2: Accuracy results for Cefadroxil

Accuracy level	Sample conc (µg/ml)	Std. conc	Total amount. Added (µg/ml)	% Recovery	Mean % Recovery	% RSD
80	15	12	27	98.47		
100	15	15	30	99.18	99.26	0.8443
120	15	18	33	100.14		

Table 3: Precision results for Cefadroxil

Parameter	Intra day	Inter-day
Sample Sol Conc.	20	20
AUC (mean +-S.D)	0.0427	0.0489
%RSD	0.9734	0.5142

Linearity and Range

The linearity was determined by using working standard solutions between 05-25 µg/ml. The areas under curve (AUC) of these solutions were recorded. Calibration curve of area under curve to concentration plotted on excel sheet and linear regression was performed. The correlation coefficient, regration Equation was calculated results were shown in Fig. 3.

Limit of Detection and Limit of Quantification

The Limit of Detection (LOD) is the smallest concentration of the analyte that gives the measurable response. LOD was calculated using the following formula

$$LOD = 3.3\ \sigma\ /S$$

.

The Limit of Quantification (LOQ) is the smallest concentration of the analyte, which gives response that can be accurately quantified. LOQ was calculated using the following formula

$$LOQ = 10\ \sigma/S$$

Where, σ is standard deviation of the response and
        S is the slope of the calibration curve.
LOD& LOQ of Cefadroxil was found to be 1.04µg/ml &3.15µg/ml respectively.

Five sets of known concentrations (05-25µg/ml) were prepared and scanned. By using these spectras, regression equations were obtained. By taking average of slopes and standard deviation of y-intercept, LOD and LOQ were calculated. The values of LOD and LOQ are given in table 4.
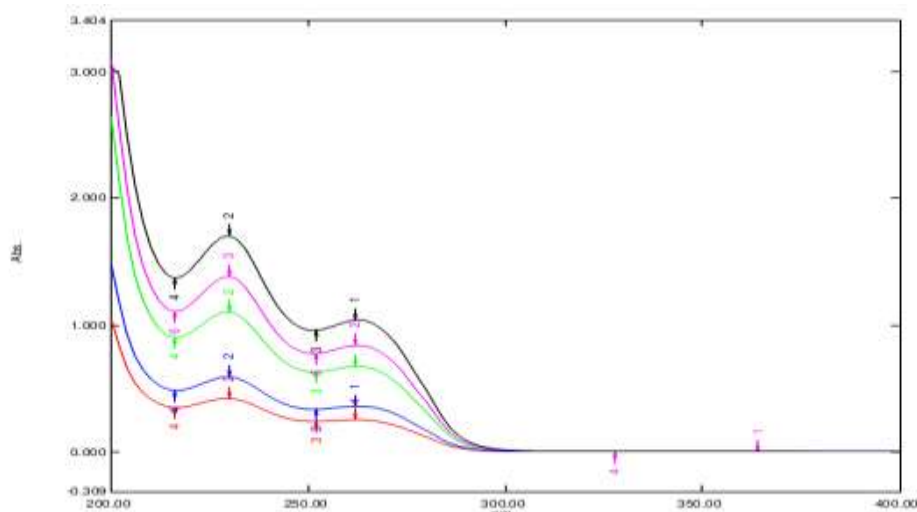
Fig 5: Overlay of Cefadroxil spectra at diff. Concentration

Table 4: Summary of validation parameters

Parameter	Result
λ range	260-266nm
Regression Equation (y=mx+c)	Y = 0.0024x0.0001
Measured wavelength	264nm
Linearity range	5-25µg/ml
Slope	0.0024
Intercept	0.0001
Correlation coefficient (R^2)	0.9993
Limit of Detection (LOD) µg/ml	1.04
Limit of Quantitation (LOQ) µg/ml	3.15
Accuracy (Mean % Recovery)	99.26
Precission (%RSD)	0.9734

RESULTS AND DISCUSSION

The UV visible spectroscopic method for the Cefadroxil by area under curve was found to be simple, accurate, economical and reproducible. The drug concentrations were found to be linear in the range of 05-25 µg/ml and the correlation coefficient value of 0.9993 indicates that developed method was linear. For Precision the percent relative standard deviation (% RSD) was found to be 0.9734 while, intra-day and inter-day precision results in terms of percent relative standard deviation values were found to be 0.9734 and 0.5142 respectively thus the method is observed as precise. The accuracy of the method was assessed by recovery studies at three different levels i.e. 80%, 100%, 120%. The values of standard deviation were satisfactory and the recovery studies were close to 100%. The % RSD value is ≤ 2 indicates the accuracy of the method. The Limit of Detection and Limit of Quantitation values were found to be1.04µg/ml &3.15µg/ml respectively. The result of the analysis for pharmaceutical formulation by the developed method was consistent with the label claim,

highly reproducible and reliable. The validation parameters are summarized in Table 4. The method can be used for routine quality control analysis of Cefadroxil in bulk and pharmaceutical formulations.

CONCLUSION

The UV spectroscopic AUC method for the analysis of Cefadroxilwas found to be simple, precise, and accurate; can be used for assay of bulk drug and pharmaceutical dosage formulations.

REFERENCES

1. Maryadele. J. O' Neil. The Merck Index: An Encyclopedia of chemicals, drugs andbiologicals, 14th edition. New Jersey: Published by Merck Research Laboratories, Division of Merck and Co., Inc. Whitehouse station: 2006. p. 313.
2. Sweetman SC, Eds. In The Martindale: The Complete Drug Reference. 35th ed. London, UK:

Pharmaceutical Press; 2007. p. 194.

3. The Indian Pharmacopoeia, Vol. II, Government of India, Delhi: The Controller of Publications; 2010. p. 998.

4. British Pharmacopoeia, Vol. I, Stationary Office, London: Medicines and Healthcare Products Regulatory Agency, 2010. p. 395.

5. The United States Pharmacopoeia, USP 32, NF 27, Vol. 2, Rockville, MD: The United States Pharmacopoeial Convention, Inc., 2009. p. 1819.

6. Thongpoon C, Liawruangrath B, Liawruangrath S, Alan Wheatley R, Townshend A. Flow injection chemiluminescence determination of cefadroxil using potassium permanganate and formaldehyde system. J Pharm Biomed Anal, 2006; 42:277-282.

7. Saleh GA. Two selective spectrophotometric methods for the determination of amoxicillin and cefadroxil. Analyst,1996; 121:641-645.

8. Chilukuri SP, Rama Rao SK, Prasad DS. Determination of cefadroxil by three simple spectrophotometric methods using oxidative coupling reactions. MicrochimicaActa, 1997; 126:167-172.

9. Ibrahim KE, El-Obeid HA. Spectrophotometric determination of cefadroxil in bulk and dosage form using sodium hydroxide. E J Chem, 2011; 8:1314-1322.

10. Aswani Kumar C, Gurupadayya BM, NavyaSloka S, Chandan RS, ThejaswiniJC. Colorimetric determination of cefadroxil and ceftriazone in pharmaceutical dosage forms. Tropical J Pharm Res, 2011; 10:81-88.

11. McAteer JA, Hiltke MF, Silber BM, Faulkner RD. Liquid-chromatographic determination of five orally active cephalosporins--cefixime, cefaclor, cefadroxil, cephalexin, and cephradine--in human serum. ClinChem, 1987;33:1788-1790.

12. Nahata MC, Jackson DS. Liquid chromatographic method for the determination of cefadroxil in its suspension and in serum. J LiqChromatogr, 1990; 13:1651- 1656.

13. Patil SD, Patil BF, Patil SP, Tarade VA, Zahid AMJ. Reverse phase HPLC method for the analysis of cefadroxil in pharmaceutical dosage form. Int J Pharm Res Dev, 2011; 3:155-160.

14. International Conference on Harmonization (ICH) of Technical Requirements for the registration of Pharmaceuticals for Human use, Validation of Analytical Procedures Methodology; ICH-Q2(R1), Geneva, 1996; pp.1-8.