INTERNATIONAL JOURNAL OF RESEARCH IN PHARMACY AND CHEMISTRY

Available online at www.ijrpc.com

Research Article

DOI: https://dx.doi.org/10.33289/IJRPC.30.5.2023.13(46)

SPECTROPHOTOMETRIC DETERMINATION OF DICLOXACILLIN BY MBTH IN PURE AND PHARMACEUTICAL DOSAGE FORM

Archana Siddabathuni*, Pravallika Challagalla, Samyuktha Nimmakuri,

Raja Rao Pokala and Padmalatha Kantamaneni

Vijaya Institute of Pharmaceutical Sciences for Women, Enikepadu,
Vijayawada - 521108, Andhra Pradesh, India.

ABSTRACT

A simple and sensitive visible spectrophotometric method is described for the assay of Dicloxacillin in pure and pharmaceutical dosage form. The method involves oxidative coupling of Dicloxacillin with 3-methylbenzthiazolinone-2-hydrazone (MBTH) reagent in the presence of ferric chloride dissolved in methanol and distilled water, apple green chromogen is formed. The complex chromogen exhibits absorption maxima at 595 nm. Regression analysis of Beer-Lambert plots showed good correlation in the concentration ranges 10-80 µg/ml. Regression coefficient was found to be 0.998. The method has been statistically evaluated and was found to be precise and accurate. The proposed method has been successfully applied to the analysis of Dicloxacillin in bulk drug and its tablet dosage form.

Keywords: Dicloxacillin, 3-methyl-2-benzothiazolinone hydrazone, ferric chloride, validation.

INTRODUCTION

Dicloxacillin is a penicillin-type antibiotic. It is used to treat different types of infections caused by bacteria such as bronchitis, pneumonia or staphylococcal infections[1]. Chemically Dicloxacillin is (2*S*,5*R*,6*R*)-6-[[3-(2,6-dichlorophenyl)-5-methyl-1,2-oxazole-4-carbonyl] amino]-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylic acid (Figure 1). Dicloxacillin exerts a bactericidal action against penicillin-susceptible microorganisms during the state of active multiplication and inhibit the biosynthesis of the bacterial cell wall[2]. Literature survey reveals that very few analytical spectrophotometric[3,4], LC-MS[5] and HPTLC[6] methods have been proposed for the estimation of Dicloxacillin in bulk and pharmaceutical dosage form. But till now there is no visible spectrophotometric method has been developed for the determination of Dicloxacillin with 3-methylbenzthiazolinone-2-hydrazone (MBTH) reagent using ferric chloride as coupling reagent. Hence, an attempt has been made to develop simple, rapid and novel spectrophotometric method for the estimation of Dicloxacillin in bulk and pharmaceutical formulation and the proposed method has been validated as per ICH guidelines[7].

MATERIALS AND METHODS

Instruments

Elico double beam UV-Visible spectrophotometer (SL-210) with 1 cm matched quartz cells, electronic dona balance were used for all spectral measurements.

Reagents
All the reagents used were of analytical grade and the solutions were freshly prepared. The reagents used in this method are triple distilled water, methanol, 0.1 N HCl, MBTH (0.3%) and $FeCl_3$ (1%).

Preparation of stock solution
The solution 1 mg/ml (1000 µg/ml) was prepared by dissolving accurately about 0.1 g of drug in 100 ml of distilled water.

Preparation of MBTH reagent
0.3 gm of MBTH reagent was dissolved in 100 ml distilled water and made up to the volume to 100 ml.

Preparation of ferric chloride solution
1 gm of $FeCl_3$ was dissolved in 0.1 N hydrochloric acid and made up to the 100 ml with 0.1 N hydrochloric acid.

Procedure
Into a series of 10 ml volumetric flasks, 1 ml of MBTH (0.3%) and 0.8 ml $FeCl_3$ (1%) and 0.1-0.8 ml of working standard solution (1 mg/ml) were added separately. The absorbance of apple green chromogen was measured at 595 nm against reagent blank. The amount of Dicloxacillin present in the sample solution was computed from its calibration curve. The calibration curve was plotted by taking concentration of drug Dicloxacillin on x-axis and absorbance on y-axis and was shown in Figure 2. The absorption spectrum was shown in Figure 3.

Procedure for assay of Dicloxacillin in pharmaceutical dosage forms
20 capsules of commercial samples of Dicloxacillin were accurately weighed and average capsule mass was calculated. To a fine powder the amount equivalent to 50 mg of Dicloxacillin was weighed and made up to 50 ml with distilled water. The solution was filtered and subjected to recommended procedure for the determination.

Validation parameters
Linearity
The calibration curve was constructed in each case by considering the absorbance measured at eight concentration levels of Dicloxacillin in distilled water (10-80 µg/ml) using the method of least squares, a line of best fit was taken and the correlation coefficient, slope and y-intercept was calculated. The amount of drug was computed either from calibration curve or from regression equation, the results are given in Table 1.

Accuracy
The accuracy of the method was determined by taking aliquots containing known quantity of Dicloxacillin and analyzed by proposed method; the results are compared with results of reference method & tabulated in the Table 2.

Precision
The precision of the method was studied by measuring 6 replications of sample containing 20 µg/ml of Dicloxacillin. The %RSD & SD was calculated and presented in Table 3.

Ruggedness
It is a measure of reproducibility of test results under normal expected operational condition from instrument to instrument & from analyst to analyst were reported in Table 4 & 5.

Specificity
Absorbance of blank solution was measured and is found to be very negligible 0.0012 hence no interference with blank solution was observed. This indicates that the method for the drug is specific.

Robustness
The robustness of the method was followed by optimizing conditions with slight variation i.e. by altering detection wavelength (1 nm) the results were reported in Table 6.

RESULTS AND DISCUSSION
The presence of amino group in Dicloxacillin enabled oxidative coupling with MBTH and $FeCl_3$ to form green colored chromogen in this method exhibiting λmax at 595 nm. The Beer's law was obeyed for this method and was linear in the concentration range of 10-80 µg/ml respectively. Regression coefficient was found to be 0.998. Hence the method showed good linearity within the range. The percent recovery for Dicloxacillin was found to be 99-100.3%. %Recovery of the drug was found to be NLT 98%-NMT 102%. Hence the method is accurate. To evaluate the validity and reproducibility of the method, known amount of pure drug was added to the previously analyzed pharmaceutical preparation and the mixtures were analyzed by the proposed method. %RSD for absorbance of 6 solutions of same concentration was found to

be 0.6127. This indicates reproducibility of the method. Hence the method is precise. No interference with blank solution was observed. This indicates that the method is specific for the drug. Analyst and instrument variation was checked. Results were within the limits. This indicates that the method is rugged. On slight variation of the method parameters i.e. wavelength, the results were not much affected. %RSD was found to be less than 2%. Hence the method is robust. The present method involves the formation of highly stable colored species which makes it easier for the determination of Dicloxacillin from pharmaceutical dosage forms in a routine manner.

CONCLUSION

The developed method is simple, rapid, selective, economical and exhibits a fair degree of precision and accuracy. The method does not involve any critical reaction conditions. The proposed spectrophotometric method can serve as an alternative method for the routine analysis of Dicloxacillin in pure drug and in pharmaceutical formulations.

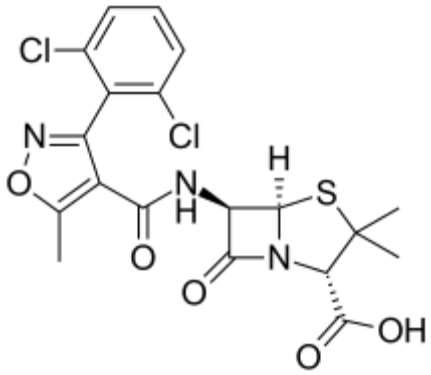

Fig. 1: Chemical structure of Dicloxacillin

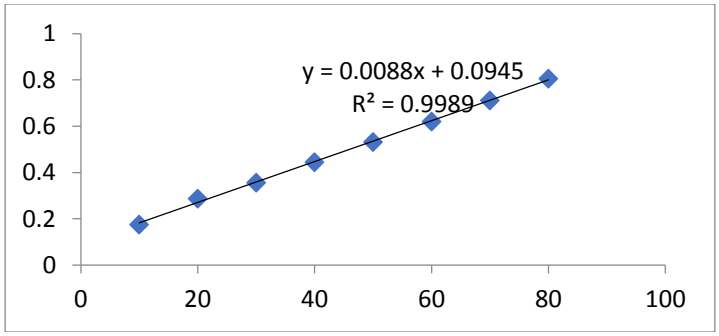

Fig. 2: Linearity curve of Dicloxacillin

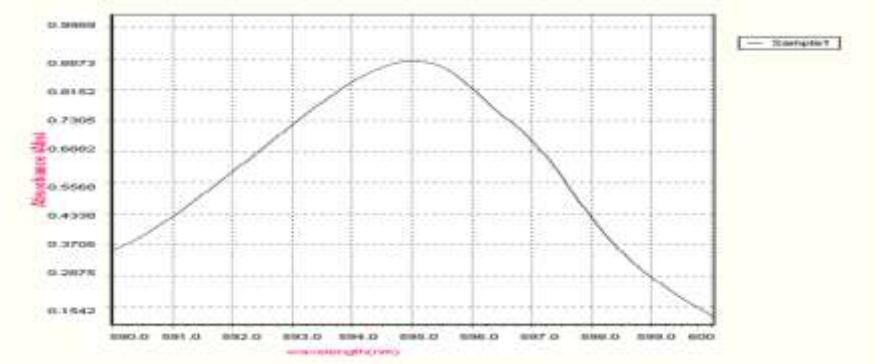

Fig. 3: Absorption spectrum of Dicloxacillin

Table 1: Linearity Results

S. No.	Concentration (µg/ml)	Absorbance
1	10	0.176
2	20	0.287
3	30	0.356
4	40	0.445
5	50	0.532
6	60	0.621
7	70	0.712
8	80	0.806

Table 2: Accuracy

Spike level`	Amount taken (µg/ml)	Amount recovered (µg/ml)	% Recovered
80%	40	40.12	100.3%
100%	50	49.89	99.7%
120%	60	59.42	99%

Table 3: Precision

Concentration (µg/ml)	Absorbance
40	0.453
40	0.457
40	0.412
40	0.433
40	0.45
40	0.454
Average	0.44316667
Std. deviation	0.01747474
% RSD	0.5461

Table 4: Instrument variation

Concentration (µg/ml)	Instrumental-1	Instrumental-2
40	0.452	0.453
40	0.451	0.452
40	0.453	0.453
40	0.453	0.452
40	0.452	0.451
Average	0.4522	0.4522
Std. deviation	0.000837	0.000837
% RSD	0.185	0.185

Table 5: Analyst variation

Concentration (µg/ml)	Analyst -1	Analyst-2
40	0.453	0.441
40	0.452	0.442
40	0.453	0.445
40	0.452	0.443
40	0.451	0.441
Average	0.4522	0.4522
Std. deviation	0.000837	0.000837
% RSD	0.187	0.185

Table 6: Robustness

Wavelength (nm)	Absorbance		
594	0.341	0.453	0.411
595	0.344	0.451	0.392
596	0.321	0.452	0.388
Average	0.3353	0.452	0.397
Std. deviation	0.0125	0.001	0.012
% RSD	3.73	0.221	0.25

REFERENCES

1. Goodman and Gilman's The Pharmacological Basis of Therapeutics. 11th ed. New York: McGraw-HILL Medical publishing division. 2005, pp. 1129.
2. The International Pharmacopoeia. 6th ed. World Health Organization. 2016, pp. 1-2.
3. Jothieswari D, Durga Prasad D, Sireesha B, Tejeswi G, Lavanya M. New analytical method for the quantitative estimation of Dicloxacillin sodium as an active pharmaceutical ingredient and in capsule dosage form. International Journal of Pharmacy & Therapeutics. 2017; 8(2): 70-75.
4. Vandana P, Angadi SS, Kale SH, Patil MA. Development and validation of stability indicating UV spectrophotometric method for the estimation of Dicloxacillin sodium in bulk drugs. Asian Journal of Research in Chemistry and Pharmaceutical Sciences. 2019; 7(1): 324-332.
5. Raj TJS, Bharati CH, Ranga Rao K, Satyanarayana Rao P, Narayan GKASS, Parikh K. Identification and characterization of degradation products of Dicloxacillin in bulk drug and pharmaceutical dosage forms. Journal of Pharmaceutical and Biomedical Analysis. 2007; 43(4): 1471-1475.
6. Prashant UT, Madhura VD, Mrinalini CD, Ashwini RM. Validated HPTLC method for determination of Dicloxacillin in simulated urine. Journal of Chemical and Pharmaceutical Research. 2013; 5(9): 77-83.
7. ICH Harmonised Tripartite Guideline. Validation of analytical procedures: Text and methodology, Q2(R1). International Conference on Harmonization. 2005; 1-13.