Volume 5 Issue VI, June 2017
ISSN: 2321-9653
International Journal for Research in Applied Science & Engineering Technology (IJRASET)

Spectrophotometric Methods for the Estimation of Third and Fourth Generation Cephalosporins In Dosage Form

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Abstract: ***The proposed spectrophotometric methods are rapid, sensitive and reproducible for the analysis of cefdinir(A), and cefepime(B) in pharmaceutical dosage forms with short analysis time. The authors developed reagent for the estimation of cefdinir and cefepime, third and fourth generation cephalosporins respectively using catechol (or p amino aceto phenone-AAP) and sodium meta periodate at a λmax of 460 nm. The Beers law range, molar absorptivity and Sandell sensitivity will be 50-250 µg/mL,4.5X10^3 and 0.03 for cefdinir respectively. In case of cefepime the Beers law range, molar absorptivity and Sandell sensitivity will be 20-250µg/mL,6.05x10^3 and 0.02 respectively.***

Keywords- Catechol , p-Amino aceto phenone and Sodium meta per iodate,cefdinir,cefepime

I. INTRODUCTION

A. Cefdinir

is chemically known as 8-[2-(2-amino-1,3-thiazol-4- yl)-1-hydroxy-2- nitroso-ethenyl] amino- 4-ethenyl-7-oxo- 2-thia-6- azabicyclo [4.2.0]oct-4-ene-5-carboxylic acid, a semi-synthetic, broad- spectrum antibiotic in the third generation of the cephalosporin class. It is used to control for common bacterial infections of the ear, sinus, throat, and skin. Therapeutic uses of cefdinir include otitis media, soft tissue infections, and respiratory tract infections, including sinusitis, strep throat, community-acquired pneumonia and acute exacerbations of bronchitis.

B. Cefepime

is chemically known as (6R,7R,Z)-7-(2-(2-aminothiozyl-4-yl)-2-(methoxyimino)acetamido)-3-((1-ethylpyrrolidinium1-yl)methyl)-8-oxo-5-thia-1-aza-bicyclo(4,2,0) oct-2-ene-2- carboxylate (Fig-1.2.1) is a fourth generation cephalosporin used to control both gram positive and gram negative bacteria. Cefepime is usually reserved to treat severe nosocomialpneumonia, infections caused by multi-resistant microorganisms (e.g. pseudomonas aeruginosa) and empirical treatment of febril eneutropenia. Cefepime has good activity against important pathogens including pseudomonas aeruginosa, Staphylococcus aureus, and multiple drug resistant Streptococcus pneumoniae. It acts against enterobacteriaceae, whereas other cephalosporins are degraded by many plasmid- and chromosome-mediated beta-lactamases.
Side effects of cefepime are mild pain, redness, or swelling at the injection site.

II. EXPERIMENTAL

A. Preparation of Solutions

Catechol : 0.1% solution was prepared by dissolving 0.1 g of catechol sample (A.R .grade SDFCL Mumbai) in 100 mL of distilled water.

Sodium meta per iodate, $NaIO_4$: 2.1392 g of NaIO4 (AR grade Hi Media laboratories Mumbai-66) was dissolved in distilled water and the total volume was brought to 1 Lt (0.01M) in a standard volumetric flask.

Standard solution of cefdinir and cefepime (in dosage form): Drug (RTIST – DT for Cefdinir or Novapime for cefepime – Lupin laboratories, Mumbai) solution was prepared by dissolving 100 mg of drug sample in 100mL of distilled water. Working solutions of drug sample (100mg / mL) were prepared by diluting aliquots of the stock solutions with distilled water.

B. Instrumentation

Spectral measurements and absorbance readings were made on Elico SL 177 double beam Spectrophotometer. pH measurements

Volume 5 Issue VI, June 2017
ISSN: 2321-9653
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were carried out using Elico pH meter model LI 615.

C. Establishment of Optimum Conditions

The optimum conditions were established in each case basing on the development of maximum color and its stability and results are presented inTable – I. Among the various oxidizing agents tried, IO4- is the best one, followed by H_2O_2. The other oxidizing agents such as IO_3^-, Fe(III), MnO_4^-, Ocl^- $Fe(CN)_6^{3-}$, were found not suitable. The efficiency of the oxidizing agent depends upon its relative reactive tendency towards reactants, (drug, catechol) products (indo-dyes) and also on the behavior of its reduced form. The formation of colored species of same λmax in the case of cefdinir (cefepime) with each pair of reagents, (Catechol – IO_4^- and AAP-IO_4^-) suggests that the indo dye formed with both compounds is the same.

Table-I Experimental Conditions

S.No	Drug	P^H	Catechol	$NaIO_4$	Maximum Colour development	Stability	λmax
1	Cefdinir	4±0.4	1 mL	1mL	5 min.	120 min.	460 nm
2	Cefepime	4±0.4	1 mL	1mL	5 min.	180 min.	460 nm

D. Assay Procedure

1) *For Cefdinir (or Cefepime) using Catechol- IO_4^-*: 15 mL of buffer solution, 0.4 –5mL of aliquots of cefdinir (Cefepime) solution,1mLof IO_4^-, 1mLof catechol(or AAP)were successively placed in a 25 ml volumetric flask, so as to make total volume of 25 mL.The absorbance of colored species was measured at 460 nm between 5—70 min. against corresponding reagent blank prepared in a similar manner. The amount of cefdinir (or cefepime) was read from calibration curve prepared with the standard solution under identical conditions.

E. Optical Characteristics

1) *Adherence to Beer's law.*: In order to test whether the cefdinir-catechol-IO_4^-(or AAP- IO_4^-) or cefepime- catechol-IO_4^- (or AAP- IO_4^-) systems adhere to Beer's law, the absorbance at λmax of a set of solutions (25mL) containing varying amounts of cefdinir (cefepime),15 mL of buffer solutions, specified concentrations of catechol, (or AAP) and oxidizing agent (Table-II) were measured against blank on spectrophotometer. The linearity of the plot between absorbance and the concentration range specified in Table-II shows that the color system obeys Beer's law, Fig's 1.1 and 1.2. Beer's limits, molar absorptivity, optimum photometric range and Sandal's sensitivity values of the method in the case of cefdinir were calculated and results are incorporated in Table-II

Table-II Optical Characteristics

Reagent	Beers Law Range µg/mL	Molar Absorptivity Lt/mol/cm	Sandell's sensitivity µg/cm2/0.001 absorbance units	Optimum photometric range µg/25 ml
Catechol-IO_4^- and cefdinir	50 – 250	4.5×10^3	0.03	100—257
Catechol-IO_4^- and cefepime	20 - 250	6.05×10^3	0.02	80 – 316

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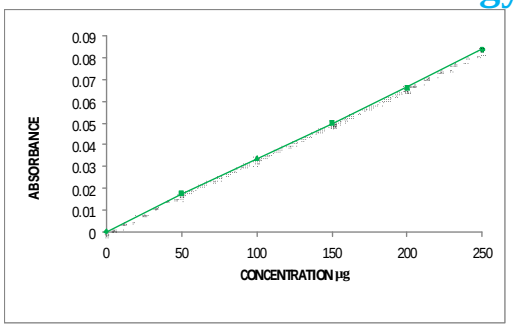
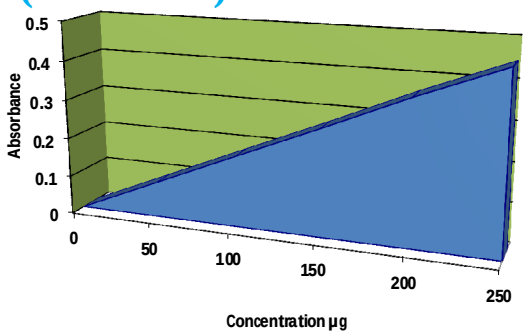


Fig.1.1 Linearity of Cefepime



(3D) Fig.1.2 Linearity of Cefdinir

F. Precision and Accuracy

The precision and accuracy of the methods in the determination of cefdinir and cefepime were tested by measuring the absorbance of six replicates each containing approximately 3/4 th of the Beer's law limit concentrations. The percentage of relative standard deviations and confidence limits (0.05 and 0.01 levels) in the methods are presented below.

Table. III. Precision and Accuracy

Cefdinir	Amount of Drug *		% Error	% R.S.D	% Range of Error	
	Taken mg	Found mg			95% Confidence Limit	99% Confidence Limit
Catechol IO4- reagent and cefdiir	0.25	0.247	1.174	2.657	±1.6	±2.92
	0.15	0.148	1.3	1.26	±1.32	±2.92
Catechol-IO_4^-reagent cefipime	0.20	0.197	1.172	1.27	±1.3	±1.98
	0.25	0.247	1.174	2.655	±1.6	±2.92

To justify the suitability of the proposed methods, known amounts of Cefdinir (or cefepime) was added to the quantities of previously analyzed samples (in dry form) and then analyzed again. The results are incorporated in Table-IV.

Table-IV. Results of analysis of formulations and Recovery experiments

Sample	Labeled Amount	Amount found	%Recovery
Ciplacef	125 mg	124.86 mg	99.88
Cefast	125 mg	124.55 mg	99.64
Cepime	500 mg	499.6 mg	99.92
Megapime	500 mg	495.75 mg	99.15

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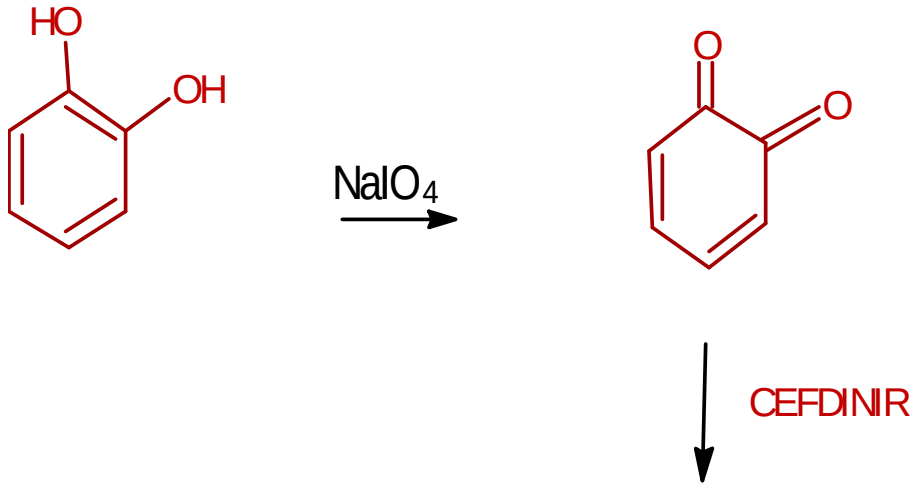
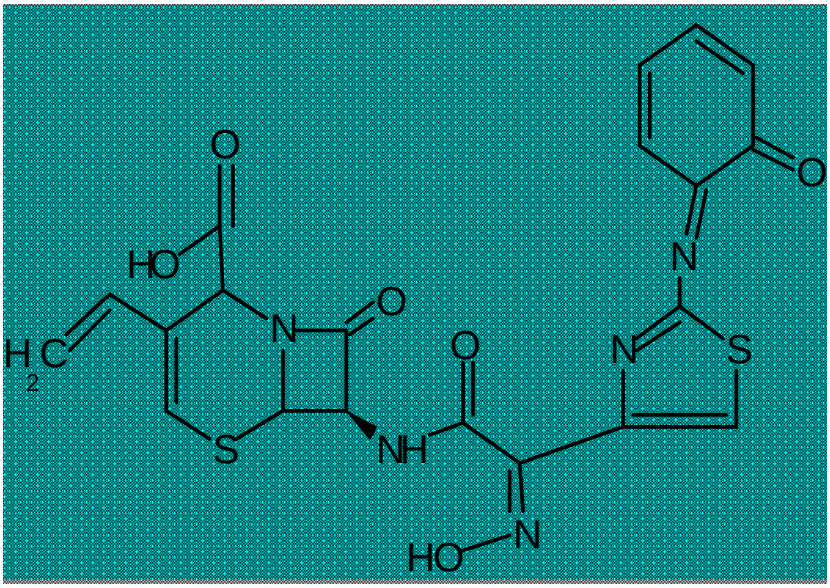
III. RESULTS AND DISCUSSION

Based on the results furnished in Tables I. –IV it can be inferred that the method proposed for the spectrophotometric determination of cefdinir (cefepime) is simple, rapid, sensitive and specific with reasonable precision and accuracy. The methods have been extended to determine cefdinir (or cefepime) different pharmaceutical preparations.

The proposed method appears to be superior to many of the reported methods7-9 and so they can be employed in routine determinations. Catechol is readily oxidisable by sodium meta per iodate to form o-benzoquinone. Cefdinir (cefepime) by virtue of its strong electron donating group, (-NH_2) oxidative coupling reaction takes place with electron deficient o-benzo quinone to form indo dye.





IV. CONCLUSION

Hence the authors conclude that the proposed spectrophotometric methods are sensitive and reproducible for the analysis of cefdinir (cefepime) in pharmaceutical dosage forms with short analysis time.

V. ACKNOWLEDGEMENTS

The authors express their sincere thanks to Prof.B.Syama Sundar former Vice chancellor of Yogi Vemana University,Kadapa A.P for his valuable suggestions and continuous encouragement.

Volume 5 Issue VI, June 2017
ISSN: 2321-9653

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