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Novel, Simple, Precise RP-HPLC Method for the Estimation of Cefotaxime Sodium in Pure and Pharmaceutical Formulations

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Abstract

The main objective of this study is to develop and validate a novel, simple, new, fast, sensitive, precise and accurate RP-HPLC analytical methods have been established for the estimation of cefotaxime sodium in bulk and pharmaceutical dosage forms.

The present method was developed and validated on a Waters HPLC system using Phenomenex make Shimadzu C18 column (250mm × 4.6mm i.d., 5µm particle size) column was used for the separation. Best results were obtained with the mobile phase composition consisting of Acetonitrile water (70:30, v/v). The system was regulated at 1.0 ml/min flow rate at 233nm UV detection.

Cefotaxime sodium was eluted at 2.622 min retention time. The analytical parameters such as accuracy, precision, linearity, LOD, LOQ, ruggedness, and robustness were used for validating the developed method according to ICH guidelines. Linearity was exhibited over the concentration range of 0.01-0.07µg/ml and the Limit of Detection and Quantitation values for cefotaxime sodium were 1.8ng/ml and 5.8ng/ml respectively. The result of analysis shows that the amount of drugs present in the formulation has a very good correlation with the label claim of the formulation and % RSD will be less than 2 for all the validation parameters. Recoveries studies revealed that results within the specified limits.

The developed methods were validated for various parameters as per ICH guidelines. Hence the proposed method was found to be satisfactory and could be used for the routine analysis of cefotaxime sodium in their marketed formulation.

Keywords: Cefotaxime sodium; Analytical parameters; ICH Guidelines; RP- HPLC.

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INTRODUCTION

Cefotaxime is a third generation cephalosporin antibiotic. Like other third generation cephalosporins, it has broad spectrum activity against Gram positive and Gram negative bacteria. It is a bactericidal agent; the β-lactam ring structure of cephalosporin interferes with the synthesis of

the bacterial cell wall. They are one of the safest and the most effective broad spectrum bactericidal antimicrobial agents and therefore, they are the most frequently prescribed class of antibiotics. Cefotaxime sodium injection is used to treat certain infections caused by bacteria including pneumonia and other lower respiratory tract (lung) infections, gonorrhoea, meningitis, and other brain spinal cord infections.[1-3]

In literature survey several analytical methods like HPLC[4,5], spectrophotometric[6,7,8] and spectrofluimetric[9-11] methods have been reported on the determination of cefotaxime sodium in combination with other drugs However few methods were reported for estimation of cefotaxime sodium individually in biological and pharmaceutical formulations by using RP-HPLC method. In view of the above facts, aim to develop aRP-HPLC method for the estimation of cefotaxime sodium in both pure and pharmaceutical dosage forms.

MATERIALS AND METHODS

A) Drugs and Formulations

Pure Cefotaxime was obtained from Lupin Pharmaceuticals, Mumbai as reference samples. Cefotaxime injection was purchased from local market.

B) Reagents and Chemicals Used

- Solvent used in HPLC (HPLC Grade)
- Distilled water (HPLC Grade)
- ACN and water as mobile phase

C) Instruments Used

Chromatographic system:

The chromatographic system used to perform development and validation of this assay method was comprised of a LC-10ATvp binary pump, a SPD-20Avp UV-Visible detector and a Hamilton manual injector with 20µl loop (Shimadzu, Kyoto, Japan) connected to a multi instrument data acquisition and data processing system (LC Solution, Shimadzu).

D) Experimental

a) Selection of Mobile phase:

The pure drug of cefotaxime sodium was injected into the HPLC system and run in different solvent systems. Different mobile phases like methanol and water, acetonitrile and acetic acid, acetonitrile and water were tried in order to find the best conditions for the separation of cefotaxime sodium. It was found that acetonitrile and water (70:30) gives satisfactory results as compared to other mobile phases.

b) Preparation of Mobile phase:

The HPLC grade acetonitrile and water were mixed together by selected ratio and ultrasonicated for 10 minutes and degassed.

c) Preparation of Standard Solutions:

10 mg of cefotaxime was weighed accurately and transferred to 10 ml standard flask, add mobile phase (acetonitrile and water -70:30) for dissolving and the volume made up to 10ml with mobile phase to get a concentration of 1000 µg/ml (Stock solution A).

d) Selection of analytical wave length:

Appropriate dilutions were prepared from the standard stock solution and scanned in the spectrum mode from 200nm to 400nm.

e) Chromatographic Conditions:

Based on the above studies, the following chromatographic condition wasfinally optimized for the estimation of cefotaxime sodium in pure and dosage form (Table 1).

Table 1: Chromatographic condition for cefotaxime sodium.

Stationary phase	**Phenomenex C_{18} Column (250 mm x 4.6 mm i.d,5µm)**
Mobile phase	Solvent A:Acetonitrile& Solvent B: Water
Solvent Ratio	70 :30
Flow rate	1ml/min.
Injection volume	20µL
Detection wave length	233nm
pH	4.2
Temperature	Ambient temperature

f) Preparation of Calibration curve:

Appropriate aliquots were pipette out from the stock solution-A in to a series of 10 ml volumetric flasks. The volume was made up to the mark with Acetonitrile and water (70:30 % v/v) to get a set of solutions having the concentration range, ranging from 0.01 to 0.07µg/ml of cefotaxime sodium. 20 µl of each concentration of the drug were injected into the HPLC system and their chromatograms were recorded under the same chromatographic

conditions as described above. Peak areas were recorded for all the peaks and a standard calibration curve was plotted.

g) Preparation of Formulation Solutions:

Twenty tablets were weighed and finely powdered in a mortar. From the powdered tablet, a quantity of powder equivalent to 10 mg of cefotaxime sodium was weighed and extracted with mobile phase having acetonitrile and water (70:30) and finally made up to 10ml with mobile phase to get a concentration of 1000 µg/ml. From this, 0.1mL of sample was drawn and made up to 10mL with mobile phase (10 µg/ml). 20 µl of sample was injected into the HPLC system and their chromatogram was recorded under the same chromatographic conditions as described above. Peak area was recorded.

h) Method or Recording of chromatogram:

With the optimized chromatographic conditions mentioned above, a steady baseline for about 20 min. After the stabilization of the baseline for about 30 min., a diluent injection of the solvents used for solubilizing drug was given in duplicate. Then the standard solution was injected and chromatogram was recorded until the reproducibility of the peak areas were found satisfactory and finally 20 µL of the standard solution of cefotaxime sodium was injected and the chromatograms was recorded. Successive aliquot of 20 µL of sample solution was injected and the chromatogram was recorded.

RESULTS AND DISCUSSION

Development of Chromatographic Method

In the present work, an analytical method based on RP-HPLC method using UV detection was developed and validated for determination of Cefotaxime sodium in pure and dosage forms. The analytical conditions were selected, keeping in mind the different chemical nature of the drugs. The development trials were taken by using the sample of each component was done, by keeping them in various extreme conditions.

Phenomenex make Shimadzu C18 column (250mm × 4.6mm i.d., 5 µm particle size) column was used for the seperation. The acidic pH range was found suitable for solubility, resolution, stability, theoretical plates and peak shape of all components. For the selection of organic constituent of mobile phase, acetonitrile was chosen to reduce the longer retention time and to attain good peak shape. Best results were obtained with the mobile phase composition consisting of a mixture of Acetonitrile water (70:30, v/v) for Cefotaxime sodium. Optimize mobile phase proportion was provide good resolution.

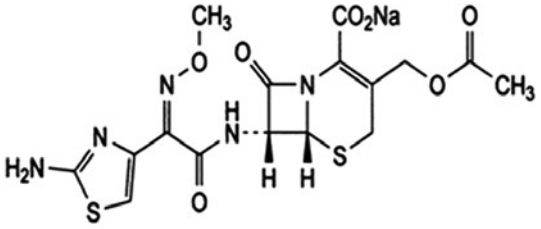

Fig. 1: Structure for Cefotaxime sodium

In this proposed method, RP-HPLC based on UV detection was developed and validated for the estimation of Cefotaxime sodium in pure and dosage forms. Phenomenex make Shimadzu C18 column (250mm × 4.6mm i.d., 5 µm particle size) column and mobile phase composition consisting of a mixture of ACN-Water (70:30v/v) found to be giving satisfactory results. For the selection of organic constituent of mobile phase, acetonitrile was chosen to reduce the longer retention time and to attain good peak shape. Fig. 2 represents the wavelength selection. Fig. 3 and Fig. 4 represent the chromatograms of standard and sample preparation respectively.

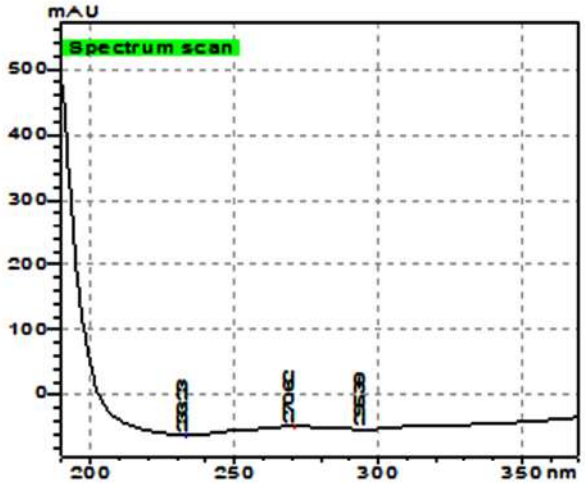

Fig. 2: HPLC Scan Spectrum for Cefotaxime sodium

Estimation

The assay procedure was repeated for 6 times, mean weight of standard drugs, of sample were taken and calculated. Prepare 10 µg/ml of standard and sample solution were also prepared and assayed for content of Cefotaxime sodium against the reference standard. The content of Cefotaxime sodium in the marketed brands was determined.

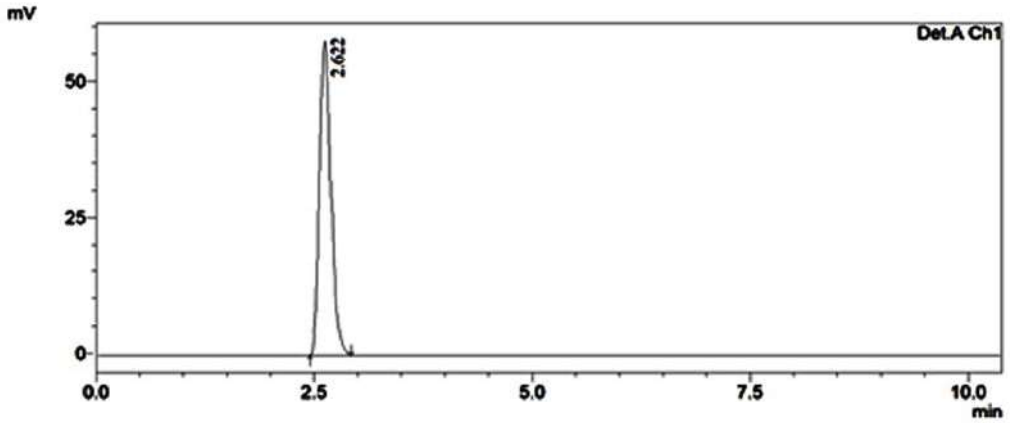

Fig. 3: Chromatogram for Cefotaxime sodium (Std)

Estimation

The assay procedure was repeated for 6 times, mean weight of standard drugs, of sample were taken and calculated. Prepare 10 µg/ml of standard and sample solution were also prepared and assayed for content of Cefotaxime sodium against the reference standard. The content of Cefotaxime sodium in the marketed brands was determined. The percentages of individual drugs found in formulations, amount and relative standard deviation in formulations were calculated. The result of analysis shows that the amount of drugs present in the formulation has a very good correlation with the label claim of the formulation (Table 2).

Table 2: Estimation of Cefotaxime sodium

Tablet Brand name	Labelled amount (g)	Estimated amount (mean±SD,g)	% Label claim	% RSD
Brand-A	0.5	0.501±0.0096	100.2%	0.019
Brand-B	0.5	0.498±0.00072	99.6%	0.014

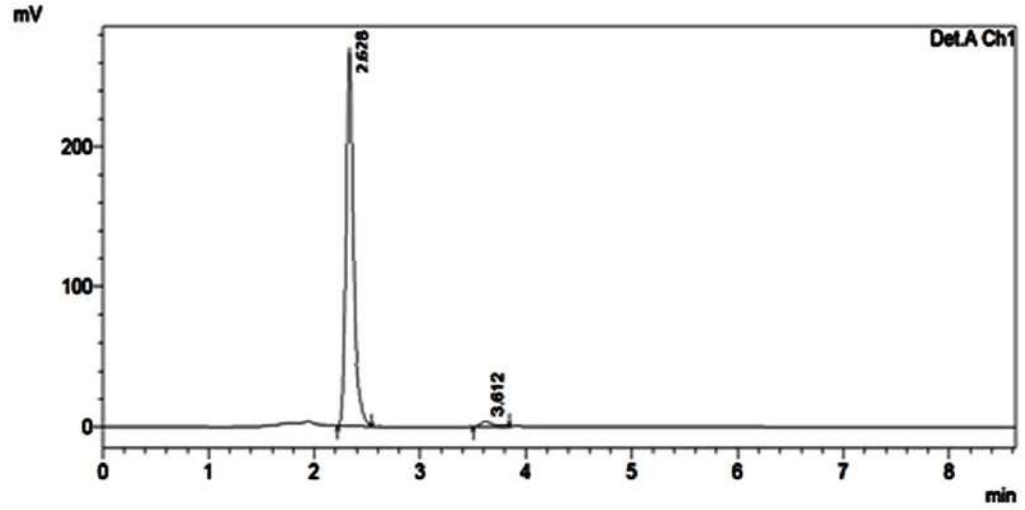

Fig. 4: Chromatogram for Cefotaxime sodium (Sample)

The chromatogram of sample as shown in Fig. 4.

METHOD VALIDATION

1) Linearity and Range

For linearity seven points calibration curve were obtained in a concentration range from 0.01-0.07 µg/ml for Cefotaxime sodium. The

response of the drug was found to be linear in the investigation concentration range and the linear regression equation for Cefotaxime sodium was y = 4E+07x+24261 with correlation coefficient 0.9995 (Fig. 5).

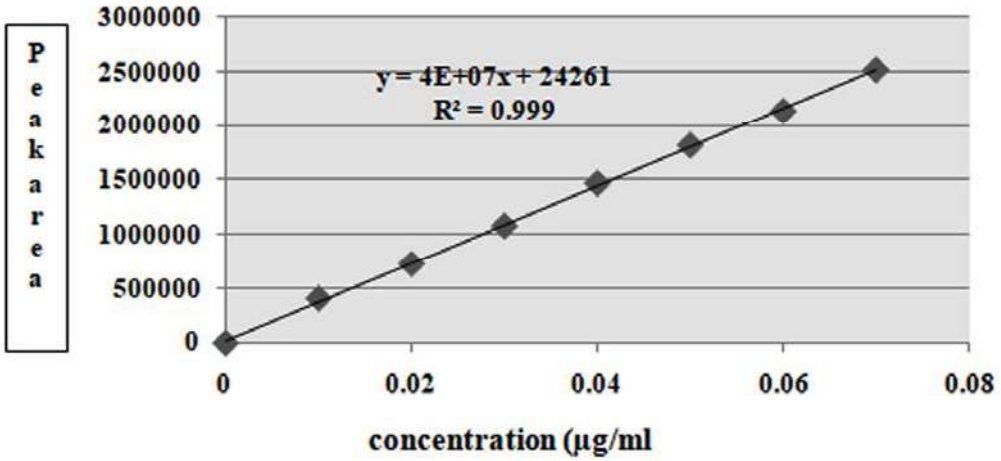

Fig. 5: Calibration curve of Cefotaxime sodium

Where x is the concentration in µg/ml and y is the peak area in absorbance unit. Chromatogram obtain during linearity study were shown in (Table 3 & 4)

Table 3: Peak area vs Concentration table of Cefotaxime sodium

Concentration (µg/ml)	Peak area
0.01	410395
0.02	731422
0.03	1083175
0.04	1474132
0.05	1820427
0.06	2141475
0.07	2521862

Table 4: Linearity Data for Cefotaxime sodium

Parameters	Data's
λmax	233 nm
Linearity (µg/ml)	0.01-0.07
Correlation coefficient (r^2)	0.9995
Slope (m)	4E+0.07
Intercept (c)	24261

2) LOD and LOQ:

The limit of detection and limit of quantification were evaluated by serial dilutions of Cefotaxime sodium stock solution. The LOD value for Cefotaxime sodium was 1.9µg/ml and LOQ was found to be 5.8g/ml. Chromatogram of LOD and LOQ study were shown in (Table 5).

Table 5: LOD and LOQ for Cefotaxime sodium

Method	Detection Wavelength (nm)	LOD ng/ mL	LOQ ng/ mL
Proposed method	233 nm	1.9	5.8

3) Accuracy:

The HPLC area responses for accuracy determination are depicted in (Table 6).

Table 6: Accuracy studies for Cefotaxime sodium

Level of addition (% pure drug)	Con. of drug in formulation (µg/ml)	Conc. drug(µg/ml) of Pure	Total conc. of drug found (µg/ml)	% Analytical recovery	Data
50%	0.05	0.025	0.075	99.46	Mean=99.463
50%	0.05	0.025	0.075	99.33	SD=0.165
50%	0.05	0.025	0.075	99.6	% RSD=0.165
100%	0.05	0.05	0.100	99.7	Mean=99.66
100%	5	0.05	0.996	99.6	SD=0.040

table cont.....

100%	0.05	0.05	0.997	99.7	% RSD=0.040
150%	0.05	0.075	0.125	99.84	Mean=99.7
150%	0.05	0.075	0.125	99.84	SD=0.075
150%	0.05	0.075	0.125	99.68	% RSD=0.075

The result shown that best recoveries (99%-101%) of the spiked drug were obtained at each added concentration, indicating that the method was accurate.

4) Precision:

The result of interday and intraday precision and repeatability of std and study are shown in (Table 7 & 8). The developed method was found to be precise as the % RSD values for the repeatability and inter and intraday precision studies were < 0.096% and < 0.268%, respectively, which confirm that method was precise.

Table 7: Precision study for Cefotaxime sodium

	Intraday (n=6)			Interday (n=6)		
Conc. (µg/ml)	**Peak area (1)**	**Peak area (2)**	**Peak area (3)**	**Peak area (D-1)**	**Peak area (D-2)**	**Peak area (D-3)**
0.05	1818432	1836524	1844587	1836524	1874587	1884587
0.05	1816542	1832542	1845625	1842542	1885625	1895625
0.05	1815628	1855988	1865584	1855988	1875584	1885584
0.05	1811325	1835699	1845262	1835699	1875262	1885262
0.05	1817458	1836647	1845636	1836647	1885636	1895636
0.05	1801547	1832566	1485698	1832566	1875698	1885698
Mean	1815324	1838754	1849845	1838754	1879845	1879845
S.D	956.28	1142	1483.2	1296	1588	1654
%RSD	0.052	0.062	0.087	0.07	0.084	0.088

Table 8: Repeatability study for Cefotaxime sodium

Conc. (µg/ml)	Peak area	Statistical data
0.05	1820427 1827482 1825317 1832615 1828513 1827482	Mean=1826870.8 SD=4472.22 %RSD=0.244

6) Ruggedness

The ruggedness of the methods was demonstrated (Table 9) by conducting the experiment on two

Table 9: Ruggedness of Cefotaxime sodium

Sl No.	Conc. (µg/ml)	Analyst-1	Analyst-2
1	0.05	1834132	1836426
2		1835284	1845146
3		1838763	1842216
4		1840284	1848127
5		1832132	18321328
6		1835482	1835282
Mean	–	18376795	1844290.5
S.D	–	3933.32	4390.2
%RSD	–	0.214	0.238

different analyst and %RSD was calculated. Variation in percentage content was found to be within the limit so the method is rugged.

7) Robustness

The result of robustness study of the developed assay method was established in (Table 10 & 11). The result shown that during all variance conditions, assay value of the test preparation solution was not affected and it was in accordance with that of actual. Hence the analytical method would be concluded as robust.

Table 10: Robustness Studies of Cefotaxime sodium (Change flow rate) (n=6)

Sl. No.	Flow rate ml/min.	Peak area	Statistical Data	Retention Time
1	1.0 ml/min.	1820423		2.628
		1834132		2.627
		1837429	Mean=1831913.4	2.628
		1836345	SD=6844.37 %RSD=0.323	2.628
		1831234		2.627
		1833598		2.629
2	0.5 ml/min.	1780312		2.624
		1783842		2.625
		1784176	Mean=1786695	2624
		1785498	SD=6331.32 %RSD=0.354	2.625
		1796823		2.626
		1788324		2.625

Table 11: Robustness studies of Cefotaxime sodium (Change in solvent concentration)

SL No:	Solvent conc. (n=6)	Peak area	Statistical Data	Retention Time
1	ACN: Water [50:50]	2428382		2.628
		2427437	Mean=100.19	2.627
		2431342	SD=1.098	2.627
		2427747	%RSD=1.096	2.628
		2425847		2.628
		2455864		2.628
2	ACN: Water [70:30] .	1834132		2.625
		1835284	Mean=100.37	2.625
		1837429	SD=0.247	2.624
		1836345	%RSD=0.229	2.625
		1831234		2.624
		1833598		2.626

8) System suitability

A system suitability test of the chromatographic system was performed before each validation run. Five replicate injections of standard preparation were injected and tailing factor, theoretical plate and % RSD of peak area were determined for same. Acceptance criteria for system suitability, tailing factor not more than 2.0, Theoretical plate not less than 5000 and % RSD of peak area not more then

2.0, were full fill during all validation parameter.

CONCLUSION

Development of methods to achieve the final goal of ensuring the quantity of drug substances and drug products is not a trivial undertaking. Cefotaxime is a third generation cephalosporin antibiotic. Like other third generation cephalosporins, it has broad spectrum activity against Gram positive and Gram negative bacteria. It is a bactericidal agent; the β-lactam ring structure of cephalosporin interferes with the synthesis of the bacterial cell wall. They are one of the safest and the most effective broad-spectrum bactericidal antimicrobial agents and therefore, they are the most frequently prescribed class of antibiotics. Cefotaxime injection is used to treat certain infections caused by bacteria including pneumonia and other lower respiratory tract (lung) infections, gonorrhoea, meningitis, and other brain spinal cord infections.[3]

In the present investigation, novel, simple, precise and accurate RP-HPLC method was successfully developed for the quantitative estimation of Cefotaxime sodium) in its pure and dosage forms by using SHIMADZU make, Phenomenex Column C18, 250 cm X 4.6 mm i.d, 5 µm particle size in isocratic mode with UV detector was used for separation and mobile phase used for the determination for cefotaxime sodium (ACN: Water - 70:30). The peak obtained were sharp retention time of 2.622 for cefotaxime sodium. Acceptance criteria for system suitability, tailing factor not more than 2.0, Theoretical plate not less than 5000 and % RSD of peak area not more then 2.0, were full fill during all validation parameter.

The assay results confirmed to the label claim of the formulation. Hence the proposed method was found to be satisfactory and could be used for the routine analysis of cefotaxime sodium in their marketed formulation. The results were found to be good and were expressed in (Table 12).

Table 12: System suitability Parameter for Cefotaxime sodium

Parameter	Cefotaxime sodium
Theoretical plate	5219
Tailing factor	0.59
Retention time	2.622
Linearity Range(µg/ml)	0.01-0.07
Regression Equation	4E+0.07x+5825.3
Slope	4E+0.07
Intercept	24216.3
Correlation Coefficient	0.9995

REFERENCES

1. Williams JD, Naber KG, Bryskier A, Hoiby N, Gould IM, Periti P, Giamarellou H, Rouveix B. International Journal of Antimicrobial Agents. 2001; 17:443.
2. Mendham J, Denney RC, Barnes JD and Thomas MK. Vogel's Text book of Quantitative Chemical Analysis. Addison Wesley Longman Ltd. 2003; 5: 1.
3. James Swarbrick, James C, Boylan, Encyclopedia of pharmaceutical technology. Marcel Dekker Inc. 1998; 217 - 224.
4. Lalitha N, Sanjay Pay PN. Development and validation of RP-HPLC method for estimation of cefotaxime sodium in marketed formultions. journal of Basic and Clinical Pharmacy. 2010; 001:67.
5. M M Aleksic, V Kapetanović, J Atanacković, B Jocić, M Zečević. Simultaneous Determination of Cefotaxime and Desacetylcefotaxime in real urine Sample usingVoltammetric and High-Performance Liquid Chromatographic Methods. Talanta. 2008; 77(1) :131-137.
6. Binglin Fan, MingjiangGeng, Yingling Wang, Quanmin Li. Spectrophotometric determination of cefotaxime by using sodium 1,2-naphthoquinone-4-sulfone. Journal of Analytical chemistry. 2013; 68: 965-968.
7. B Morelli. Derivative Spectrophotometry in the analysis of mixtures of Cefotaxime sodium and Cefadroxil monohydrate, Journal of Pharmaceutical and Biomedical Analysis. 2003; 32 (2): 257-267. Z AAlothman, M A Abdalla. Oxidative Coupling for the Spectrophotometric determination of certain Cephalosporins and acetaminophen in drug formulation.Arabian journal of chemistry. 2011; 4(2): 239-242.
8. M MAyad, AAShalaby, H E Abdellatef, H M Elsaid. Spectrophotometric determination of certain Cephalosporins through oxidation with Cerium (IV) And 1- Chlorobenzotriazole. Journal of Pharmaceutical and Biomedical Analysis. 1999; 20(3): 557-564.
9. D Chen, H Wang, Z Zhang, L Ci, X Zhang. Chemiluminescence determination of Cefotaxime Sodium with Flow-Injection Analysis of Cerium (IV)-Rhodamine 6G System and its application to the binding study of Cefotaxime sodium to protein with On-line Microdialysis sampling. SpectrochimicaActa, Part A. Molecular and Biomolecular Spectroscopy. 2011; 78(1):553-557.
10. Daniela Ştirbeț, Ioanavasilescu, Gabriel-Lucian Radu. Spectrofluorimetric analysis of cefotaxime

sodium by using 4-fluoro-7-nitrobenzofurazan as derivatization agent. U.P.B. Scientific Bulletin, Series B. 2014; 76(3): 89-98.

11. M A Omar, O H Abdelmageed, T Z Attia. Kinetic Spectrofluorimetric determination of certain Cephalosporins in Human plasma. Talanta. 2009; 77 (4): 1394-1404.
12. ICH, Q2 (R1), Validation of Analytical Procedures: Text and Methodology. 2005.
13. M S. Arayne, N Sultana, M Nawaz. Simultaneous Quantification of Cefpirome and Cetirizine or Levocetirizine in pharmaceutical formulations and human plasma by RP-HPLC. Journal Of Analytical Chemistry. 2008; 63 (9): 881-887.
14. J W Bae, C S Myung, C I Choi, S M Moon, C G Jang, S Y Lee. HPLC assay method for Ceftibuten in plasma and its application to pharmacokinetic study. Journal of Analytical Chemistry. 2010; 65 (12):1261-1265.
15. N O Can, G Altiokka, H Y Aboul-Enein. Determination of Cefuroxime axetil in tablets and biological fluids using Liquid chromatography and flow injection analysis. Analytica Chimica Acta. 2006; 576(2): 246-252.
16. R Denooz, C Charlier. Simultaneous Determination of five Beta-lactam antibiotics (Cefepim, Ceftazidim, Cefuroxim, Meropenem and Piperacillin) in Human plasma by High-Performance Liquid Chromatography With Ultraviolet Detection. Journal of Chromatography B Analytical Technologies in the Biomedical and Life Sciences. 2008; 864(2): 161-167.
17. F J J Palacios, M Callejón Mochón, J C Jiménez Sánchez, M Á Bello López, A Guiráum Pérez. Validation of a HPLC method for determination of Cefepime (A Fourth-Generation Cephalosporin) determination in human serum, cerebrospinal fluid, and urine pharmacokinetic profiles. Chromatographia. 2005; 62(7-8): 355-361.
18. Iqbal MS, Bahari MB, DarwisY, Iqbal MZ, Hayat A, Gantala V. An RP-HPLC-UV Method with SPE for Cefotaxime in All-in-One Total Parenteral Nutritional Admixtures: Application to Stability Studies. Journal of AOAC International, 2013; 96 (2):290-294.
19. Al-Hakkani MF. Forced degradation study with a developed and validated RP-HPLC method for determination of cefpodoximeproxetil in the bulk and finished pharmaceutical products. Journal of the Iranian Chemical Society. 2019; 16: 1571-1578.
20. Robinson AM, Medlicott NJ, Ussher JE. The rapid detection of cefotaxime-resistant Enterobacteriaceae by HPLC. Future science OA. 2016; 2: 142.
21. LI Jing, ZHANG Mei-Yun, LI Quan-Min. Spectrophotometric Determination of Cefotaxime Sodium with Potassium Ferricyanide. Chinese Journal of Applied Chemistry. 2011; 28:88-93.
22. J Shah, M R Jan, S Shah, Inayatullah. Spectrofluorimetric method for determination and validation of Cefixime in pharmaceutical preparations through derivatization with 2-Cyanoacetamide. Journal of Fluorescence. 2011; 21(2): 579- 585.
23. M Espiosa Bosch, A J Ruiz Sanchez, F Sanchez Rojas, C Bosch Ojeda. Recent Developments In The Analytical Determination of Cefadroxil. Asian Journal of Pharmaceutical Sciences. 2008; 3(3): 217-232.
24. M Andrasi, A Gaspar, A Klekner. Analysis of Cephalosporins in bronchial Secretions by capillary electrophoresis after simple pre-treatment. Journal of Chromatography B Analytical Technologies in the Biomedical and Life Sciences. 2007; 846(1-2): 355-358.
25. Bagheri AG, Yosefi A, Rezvani M, Roshanzamir S. Spectrophotometric complexation of cephalosporins with palladium (II) chloride in aqueous and non-aqueous solvents. Spectrochimica Acta Part A. 2012; 89:317– 321.