Trop J Nat Prod Res, October 2021; 5(10):1716-1723 **ISSN 2616-0684 (Print)**
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Original Research Article

Eco-Friendly and Cost-Effective Method for the Quantitative Determination of Ceftriaxone Sodium in Pharmaceutical Drug Formulations

Zeeshan Haider[1], Zeeshan Ali[1], Javaria Tayyab[1], Muhammad M. Ahmad[1], Muhammad Ahmad[2], Muhammad Sajid[1]*, Dilshad Hussain[3]

[1]*Institute of Chemical Sciences, Bahauddin Zakariya University, Multan, Pakistan*
[2]*Department of Chemistry, University of Sahiwal, Pakistan*
[3]*HEJ Research Institute of Chemistry, International Centre for Chemical and Biological Sciences, University of Karachi, Karachi, Pakistan*

ARTICLE INFO

Article history:
Received 01 January 2021
Revised 11 March 2021
Accepted 18 October 2021
Published online 02 November 2021

ABSTRACT

Ceftriaxone sodium is a beta-lactam antibiotic with bacteriocidal activity. This drug is eliminated in the urine, although only in a 33-67% concentration. Many High-Performance Liquid Chromatography (HPLC)-based methods for the analysis of ceftriaxone have been developed using various solvents, but these solvents are expensive, poisonous, and harmful. This study was therefore aimed at developing an eco-friendly and cost-effective HPLC-based approach for the quantitative determination of ceftriaxone sodium in pharmaceutical drug formulations. In the method development, the column used was X-Bridge C-18 (4.6 mm x 250 mm, 5 µm) for the separation, identification, and determination. The mobile phase was a mixture of phosphate buffer and methanol in the ratio of 73:27 (% v/v) at a flow rate of 1 mL/min, while the temperature was maintained at 25°C. Throughout the method development, the photodiode array detector was at a wavelength of 254 nm. The developed approach demonstrated good system applicability, with an RSD of less than 2%. R^2 was 0.9998, which is exact because the % RSD is less than 2% (repeatability with 0.5% RSD and intermediate precision with 0.22% RSD). The limit of detection (LOD) and limit of quantification (LOQ) for the method were 0.0472 and 0.157 mg/mL, respectively. The R^2 and RSD values suggested that the approach was linear, exact, and accurate, demonstrating that the developed method is environmentally friendly, cost-effective, and green.

***Keywords*:** Ceftriaxone sodium, Cost-effective, Eco-friendly, High-Performance Liquid Chromatography, Method development, Pharmaceutical drug formulation.

Introduction

Rocephin is the brand name of ceftriaxone sodium.[1] Ceftriaxone is a semi-synthetic antibacterial or antibiotic[2] injection. It is chemically known as [6R-[6a,7b, (Z)]-5-thia-1-azabicyclo-[4.2.0]-oct-2-ene-2-carboxylic acid, 7-[(2-amino-4-thiazolyl) (methoxyimino)-acetyl]amino]-8-oxo-3-[[(1,2,5,6-tetrahydro-2-methyl-5,6-dioxo-1-2,4-triazin-3-yl)-thio]methyl], in the hydrated disodium salt and contains about 83 mg of sodium per gram of ceftriaxone.[3] Ceftriaxone belongs to the third-generation Cephalosporin antibiotics.[4] It is a drug that prevents bacteria from producing muco-peptide in their cell walls.[5] A component of ceftriaxone's beta-lactam binds to endopeptidases, transpeptidases, and carboxypeptidases found in the cytoplasmic membrane of bacteria. When ceftriaxone binds to these enzymes, it causes abnormal cell wall formation and cell death. Ceftriaxone is a medication that is absorbed entirely and administered intramuscularly and intravenously. Its lengthy duration of action is due to two factors: a high fraction of protein binding in the plasma and delayed urine elimination.[6] Ceftriaxone can be found in urine at concentrations ranging from 33 to 67%.

Furthermore, some concentrations are eliminated as microbiologically inactive molecules in the bile and feces.[7] The majority of solvents and chemicals used in the pharmaceutical analysis are toxic, expensive, endangering workers' health, and the environment.[8] Green chemistry saves the environment by substituting greener solvents for harmful solvents or reducing solvent volume.[9] The pharmaceuticals and their contaminants are quantified using an environmentally friendly and cost-effective High-Performance Liquid Chromatography (HPLC) technique. Because of the method's relative standard deviation and linearity, it is a more ecologically friendly technique that saves time and solvents.[10] The solvent's profile is environmentally friendly, more efficient, and uses less solvent. A binary cocktail of antibiotics, ceftriaxone sulfate, and streptomycin sodium was analyzed by 'ratio spectra 2nd derivative' and 'zero-crossing 3rd derivative spectrophotometry'. The detection limits for ceftriaxone and streptomycin were calculated to be 0.15 and 0.27 mg/mL, respectively.[11] Tigecycline (TGC) has previously been shown to have clinical efficacy and safety in the treatment of complex intra-abdominal infection (CIAI) when compared to imipenem/cilastatin.[12] Microbiological responses were similar in the two treatment groups, with 68.1 % (94/138) and 71.5 % (98/137) of TGC-treated and ceftriaxone-onemetronidazole-treated subjects, respectively, achieving microbiological eradication at total plate count (TOC).[13] Using a conductometric approach in water and the presence of different salts, a recent study investigated the interaction among cephalosporin, ceftriaxone sodium trihydrate, a cationic surfactant, cetyl trimethyl ammonium bromide such as NaCl, Na_2SO_4, and Na_3PO_4 at various temperatures.[14] A single chamber microbial fuel cell with an air cathode has been successfully developed using glucose–ceftriaxone

Corresponding author. E mail: dr.msajid@bzu.edu.pk
Tel: +923040801998

Citation: Haider Z, Ali Z, Tayyab A, Ahmad MM, Ahmad M, Sajid M, Hussain D. Eco-Friendly and Cost-Effective Method for the Quantitative Determination of Ceftriaxone Sodium in Pharmaceutical Drug Formulations. Trop J Nat Prod Res. 2021; 5(10):1716-1723. doi.org/10.26538/tjnpr/v5i10.4

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

sodium combinations or ceftriaxone sodium as fuel.[15,16] The elimination of any drug residues from the related equipment is a crucial step in the pharmaceutical industry. To regulate a cleaning procedure, an HPLC-UV technique for determining ceftriaxone sodium residues on stainless steel surfaces was developed and validated. The quantification of ceftriaxone in the presence of statin medications in the formulation and human serum was reported using a reverse-phase high-performance liquid chromatographic (RP-HPLC) approach that was accurate, sensitive, and time-consuming.[17,18] The characterization of *in vitro* ceftriaxone sodium leads to an ideal biocompatibility recipe and cell assessment (hemolytic testing).[19,20]

For the analysis of ceftriaxone sodium, several different methods have been published, including chromatographic and spectrophotometric approaches. The purpose of developing a novel method for the analysis of ceftriaxone sodium was to create a method that is more cost-effective, reliable, efficient, and less harmful (in terms of chemical utilization) while still providing reasonable accuracy and precision. The chemicals utilized are both environmentally acceptable and cost-effective (Methanol and buffers). The development of green technology for measuring ceftriaxone at extremely short intervals is a primary goal of adopting this mobile phase.

Therefore, the aim of the current study was to develop an eco-friendly, cost-effective, reliable, efficient, and less harmful HPLC-based method for the quantitative determination of ceftriaxone sodium in pharmaceutical drug formulations.

Materials and Methods

Sources of chemicals

The chemicals utilized in this research were of analytical quality and were used without further purification. Methanol (HPLC grade) was obtained from Fisher Chemicals. Potassium dihydrogen phosphate, disodium hydrogen phosphate, and orthophosphoric acid were purchased from VWR Chemicals, while Brooks Pharma supplied the Ceftriaxone sodium (STD) and Titan-1000 injections.

Method development

Different trials were made to develop a method for the quantitative determination of ceftriaxone sodium. Guidelines for method development were taken from British Pharmacopeia.[21] Variation in the flow rate and composition of the mobile phase was made,[22] to get the optimum results of ceftriaxone sodium determination in active pharmaceutical ingredient and pharmaceutical injection dosage form,[23] as shown in Table 1. After numerous trials, a phosphate buffer and methanol mixture in a ratio of 73:27 was chosen as the mobile phase for the analysis and validation of ceftriaxone sodium. A 73:27 mixture of buffer and methanol was utilized for the HPLC analysis. The composition of the buffer for the mobile phase was: 800 mL water, 3.5 g potassium dihydrogen orthophosphate, and 11.6 g disodium hydrogen phosphate, with a pH of 7.

Table 1: Chromatographic conditions for HPLC-based method development

Column Used	Mob. Phase Composition	Flow Rate (mL/min)	Temp. (ºC)	Inj. Volume (µL)	Detection Wavelength (nm)
X-Bridge C-18	Buffer : Methanol 73: 27	0.8	25	10	254
X-Bridge C-18	Buffer: Methanol 73: 27	1	25	10	254
X-Bridge C-18	Buffer: Methanol 73: 27	1.2	25	10	254

HPLC analysis of ceftriaxone sodium

Ceftriaxone sodium (25 mg) was diluted in a 50 mL volumetric flask to a final volume of 50 mL, and a 0.04 mg/mL standard solution was produced from the stock solution. The mobile phase of buffer and methanol was used to evaluate the standard solution of ceftriaxone sodium on an AGILENT C-18 (250 mm x 4.6 mm, 5 m) analytical column (73:27). A photodiode array (PDA) detector was used to detect a wavelength of 254 nm. Because the usual solution peak occurred sooner than 5 minutes, the overall run time was estimated to be between 8 and 10 minutes.[24,25]

Method validation

The approach was validated using the British Pharmacopeia (BP), the United States Pharmacopeia (USP), and the International Conference of Harmonization (ICH) guidelines.[22] Linearity, Specificity, Range, Limit of Quantification (LOQ), Limit of Detection (LOD), Precision, and Accuracy were the metrics investigated during the validation. Concentrations of 0.01, 0.02, 0.04, 0.06, 0.08, and 0.1 mg/mL were the solution ranges utilized for the method validation. The range for this method was measured between 0.01 and 0.1 mg/mL, by using six different concentrations (0.01, 0.02, 0.04, 0.06, 0.08, and 0.1 mg/mL) of ceftriaxone sodium. Accuracy (%) was obtained by spiking 50, 100, and 150% working standards in the target concentration.

Method verification

Method verification for quantitative analysis of ceftriaxone sodium injection dosage form was performed by taking two different brands of ceftriaxone sodium accessible in the market.

Sample preparations of Ceftriaxone and Titan-1000 injections

To get a uniform sample of ceftriaxone sodium in Ceftriaxone injection, the whole content of a vial containing 1000 mg of ceftriaxone was transferred to a 100 mL volumetric flask and dissolved in water, sonicated for 2 minutes in a sonicator, and cooled at room temperature. Then, 1 mL from the above first dilution was taken into another 50 mL volumetric flask and filled up to the mark. Furthermore, 5 mL from the second dilution was taken into another 25 mL volumetric flask to prepare a final concentration of 0.04 mg/mL of ceftriaxone. The same method used for the Ceftriaxone sodium injection was also utilized for the Titan-1000 injection, for the dilution up to 0.04 mg/mL concentration of ceftriaxone.

HPLC *analysis for method v*erification

HPLC was used to separate the product samples. The sample sequence was performed using the sequence set method (SSM) on the HPLC equipment. To test the stability of the HPLC system, five replicates of a ceftriaxone sodium standard solution were run. This was followed by three replicates of sample solutions from two different manufacturers; all under identical conditions as the standard solution. The separating column and system tubing were washed to eliminate any leftover excipients.

Results and Discussion

Ceftriaxone sodium is a third-generation antibacterial agent, belonging to the Cephalosporin class. Ceftriaxone contains a 3-thiamethyl group as a highly acidic heterocyclic system. When compared to previously reported methods, the approach developed for this medicine is simple, cost-effective, and produces better results. Acetonitrile (ACN), ammonia, and methanol were commonly employed in prior procedures. ACN and ammonia are both harmful and costly. The validation of a developed method is carried out following the requirements of the British Pharmacopeia (BP), the International Conference of Harmonization (ICH), and the United States Pharmacopeia (USP).

Outcome of the method development

HPLC chromatogram of ceftriaxone sodium obtained from the developed method is shown in Figure 1. The retention time of ceftriaxone sodium was 4.06 minutes. The results obtained by this method are very precise.

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Five replicates of prepared standard solution (0.04mg/mL) were analyzed and the acquired data is given in Table 2, which shows the relative standard deviation of 0.5%. The method developed is cost-effective, precise, less time-consuming, accurate, and eco-friendly as chemicals used in this method are easily available, non-toxic to the environment, and cheap. It also increases the lifetime of a Photodiode Array detector lamp. As the retention time of the peak is about 3 to 4 minutes, solvent consumption is quite low, so the developed method is economical.

Outcome of the method validation

In this study, a green and low-cost method for the analysis of ceftriaxone sodium was developed and then validated to ensure the stability and authenticity of the results acquired by using the specifically developed method. The method was validated according to international guidelines; U.S Pharmacopeia, and International Conference of Harmonization. In the developed method, the following parameters were validated: the linearity, limit of detection, limit of quantification, range, precision, accuracy, and specificity.

Linearity

In this study, linearity was measured by analyzing six concentrations that range from 0.01 to 0.1 mg/mL (0.01, 0.02, 0.04, 0.06, 0.08, and 0.1 mg/mL) under the same conditions as presented in the method (Table 3). Also, three replicates of each concentration were analyzed to check the accuracy of the developed method. Linearity is very significant. It describes the limits both from the lower and upper end in which the targeted concentration of the substance in the sample can be detected. To verify the linearity of the obtained results, the calibration curve was drawn between the areas of chromatographic peak against concentration of analyte in the sample. The correlation coefficient is highlighted in Figure 2.

Limit of detection and limit of quantification

The limit of detection (LOD) of the analytical method is the minimum amount of analyte which can be detected by using that specific method. In this case, from all the above three methods, the last method was used which involves the intercept and standard deviation calculation, and the obtained LOD for the ceftriaxone sodium was 0.0472 mg/mL (Table 4). The process by which the minimum amount of analyte can be accurately quantitated is known as the limit of detection of the analytical procedure. Similarly, the limit of quantification for ceftriaxone sodium was 0.157 mg/mL as shown in Table 5.

Precision

A degree of closeness between observed values obtained by series of measurements of the same sample over an extended time by using the same method is known as precision. The precision of the analytical method (reproducibility, repeatability, and intermediate precision) was calculated. Reproducibility is only required when standardization of method is required by the International Conference of Harmonization, so it is an optional factor. Repeatability and intermediate precision were also calculated for the developed method.

Limit of detection and limit of quantification

The limit of detection (LOD) of the analytical method is the minimum amount of analyte which can be detected by using that specific method. In this case, from all the above three methods, the last method was used which involves the intercept and standard deviation calculation, and the obtained LOD for the ceftriaxone sodium was 0.0472 mg/mL (Table 4). The process by which the minimum amount of analyte can be accurately quantitated is known as the limit of detection of the analytical procedure. Similarly, the limit of quantification for ceftriaxone sodium was 0.157 mg/mL as shown in Table 5.

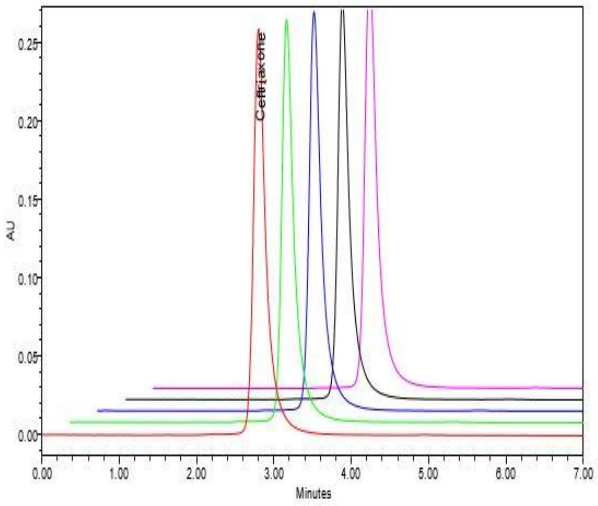

Figure 1: Chromatogram of ceftriaxone sodium standard.

Table 2: Six replicates of HPLC standard solution.

Concentration (mg/mL)	Replicate	Peak Area
0.04	1	2105635
	2	2128995
	3	2135637
	4	2131604
	5	2132577
	6	2133346
Mean		2127964.7
SD		11153.392
%RSD		0.5

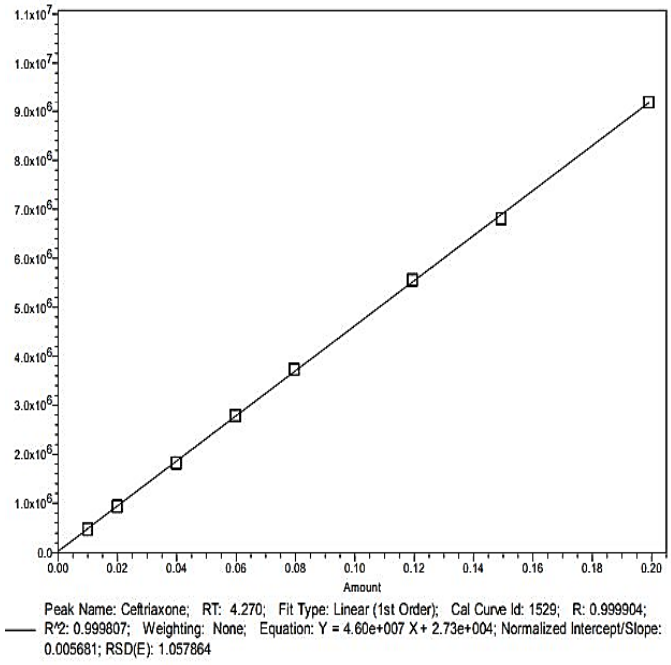

Figure 2: Linearity curve for ceftriaxone sodium

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Table 3: Linearity for developed method validation

Conc. of STD (mg/mL)	Peak Area			
	Replicate 1	**Replicate 2**	**Replicate 3**	**Average**
0.01	481789.24	478934.611	474328.476	478350.77
0.02	941282.49	934139.93	942392.92	939271.77
0.04	1823190.55	1821575.95	1816060.17	1820275.55
0.06	2786771.83	2787682.43	2791028.49	2788494.25
0.08	3735204.66	3740140.93	3730788.36	3735377.98
Correlation Coefficient (R^2)		**Intercept**	**%RSD of Mean**	**Comments**
0.9998		**-412.257**	**1.041288**	**Linear**

Table 4: Calculation of Limit of Detection

Conc. of STD (mg/mL)	Peak Area			
	Replicate 1	**Replicate 2**	**Replicate 3**	**Average**
0.01	481789.24	478934.611	474328.47	478350.77
0.02	941282.49	934139.934	942392.9	939271.77
0.04	1823190.55	1821575.95	1816060.17	1820275.55
0.06	2786771.83	2787682.43	2791028.49	2788494.25
0.08	3735204.66	3740140.93	3730788.36	3735377.98
Correlation Coefficient (R^2)		**Intercept**	**%RSD of Mean**	**Comments**
0.9998		-412.257	1.041288	Linear
Limit of Detection			0.0472 mg/mL	

Table 5: Calculation of Limit of Quantification

Conc. of STD (mg/mL)	Peak Area			
	Replicate 1	**Replicate 2**	**Replicate 3**	**Average**
0.01	481789.24	478934.61	474328.47	478350.77
0.02	941282.49	934139.93	942392.92	939271.77
0.04	1823190.55	1821575.95	1816060.17	1820275.55
0.06	2786771.83	2787682.43	2791028.49	2788494.25
0.08	3735204.66	3740140.93	3730788.36	3735377.98
Correlation Coefficient (R^2)		**Intercept**	**%RSD of Mean**	**Comments**
0.9998		-412.257	1.041288	Linear
Limit of Quantification			0.157 mg/mL	

Precision

A degree of closeness between observed values obtained by series of measurements of the same sample over an extended time by using the same method is known as precision. The precision of the analytical method (reproducibility, repeatability, and intermediate precision) was calculated. Reproducibility is only required when standardization of method is required by the International Conference of Harmonization, so it is an optional factor. Repeatability and intermediate precision were also calculated for the developed method.

Repeatability

Repeatability is the degree of closeness of results obtained by analyzing the number of replicates of the same sample under the same conditions. The repeatability of the method was studied by preparing a 0.04 mg/mL concentration solution of ceftriaxone sodium. Six replicates of 0.04 mg/mL were analyzed on the HPLC (Table 6).

Intermediate precision

According to the international guidelines, intermediate precision of an analytical procedure is the ability to produce precise results either by analyzing the same sample by a different analyst, on a different instrument, or by analyzing it on different days. In this case, intermediate precision for the analytical method was studied by testing the sample on different days. A concentration (0.04mg/mL) of solutions was prepared and ran on the same HPLC instrument but on different days. Five replicates of the standard solution were prepared on both days and analyzed on the same HPLC instrument and the results acquired by these replicates are described in Table 7. The result obtained for the square root of $\sum (\%RSD)^2$ was 0.22361. The results are within the specified limit.

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Table 6: Calculation of Repeatability

Concentration (mg/mL)	Replicates	Peak Area
0.04	1	2105635
	2	2128995
	3	2135637
	4	2131604
	5	21322577
	6	2133346
Mean		2127964.778
SD		11153.392
%RSD		0.5

Table 7: Calculation of Intermediate Precision

Concentration (mg/mL)	Replicates of STD	Day 1	Day 2
	1	892371	862750
	2	888007	864142
0.04	3	889171	863367
	4	890535	863651
	5	890720	862243
Mean		890160.815	863230.639
SD		1655.202	747.133
%RSD		0.2	0.1
$(\%RSD)^2$		0.04	0.01
Sum of $(\%RSD)^2$		0.05	
SQRT of Sum		0.22361	

Accuracy

Accuracy is the degree of closeness of the resultant values from the analytical method to the conventionally accepted reference values or true values of the analyte. In the method validation, three different concentration solutions were prepared by spiking with 50, 100, and 150% amount to the targeted concentration (0.04 mg/mL) of the sample. Three replicates of each concentration of the analyte were run on HPLC and the results acquired are presented in Table 8. The obtained recoveries for the 50, 100, and 150% spiked concentration showed recoveries up to 99.19, 98.40, and 98.48%, respectively (Table 8). These results demonstrate that the accuracy of the false method is close to each other (Figure 3).

Uncertainty

The square root of the sum of the squares of % RSD of all the parameters gives a combined uncertainty. The expanded uncertainty is shown in Table 9, while Table 10 highlights a detailed summary of the method validation.

Uncertainty

$= \sqrt{((\%RSD\ of\ Linearity)2) +}$

$(\%RSD\ of\ Bias)2\ (\%RSD\ of\ Precision)$

Method verification

After the validation studies of the developed method were achieved, it was applied to the injection dosage form for qualitative and quantitative studies of the ceftriaxone sodium by using two different pharmaceutical brands as Cefxon and Titan-1000 injections. The developed method shows no interference by excipients and additives present in the dosage form. Chromatograms of the dosage forms are shown below:

Ceftriaxone injection (1000 mg/vial)**:** The developed method was successfully applied to Cefxon injection manufactured by Bosch Pharma. The results of the chromatogram of sample solution analysis (Figure 4A) were within the range of system suitability parameters.

Titan-100 injection (1000 mg/vial): The developed method was successfully applied to Titan-1000 injection manufactured by Macter International Pharma. The results were within the range of system suitability parameters. A chromatogram of the analysis of sample solution is given in Figures 4A and B.

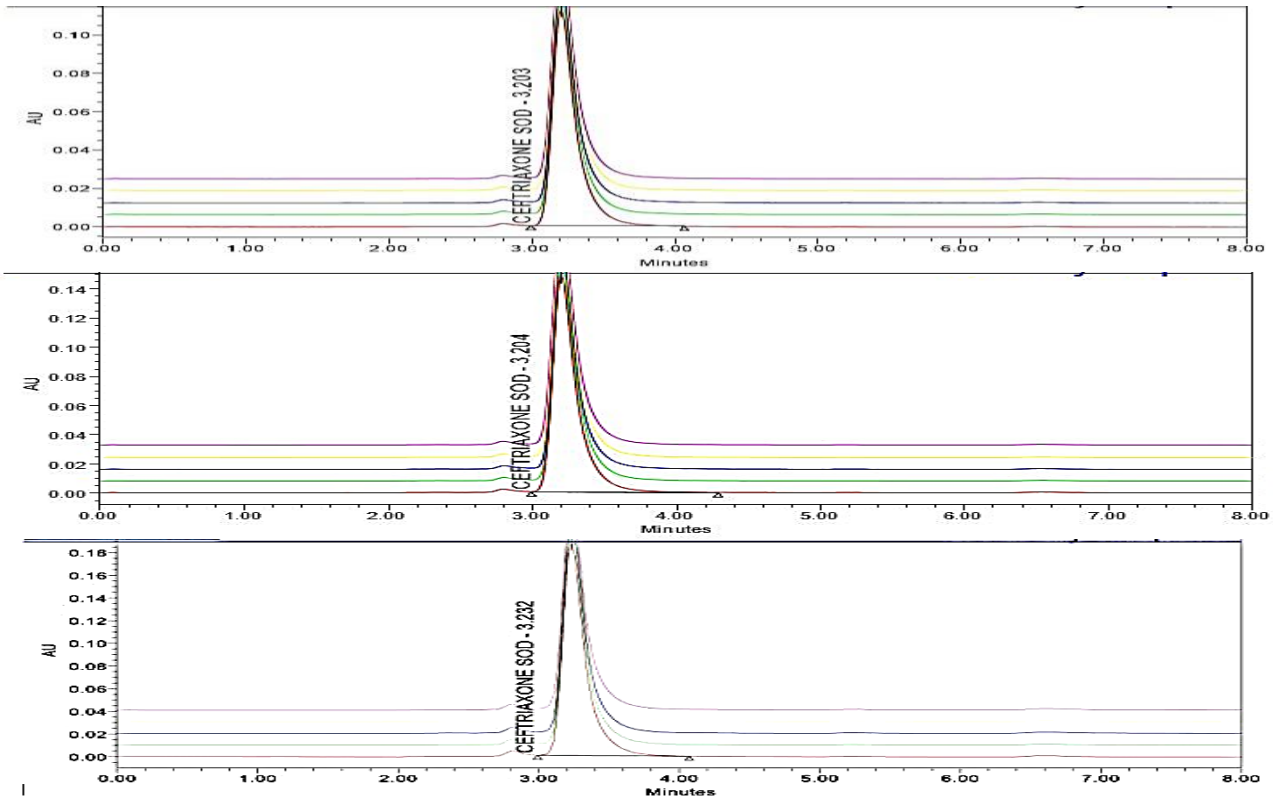

Figure 3: Chromatogram and peak response for (A) 50; (B) 100; (C) 150 % spiking

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

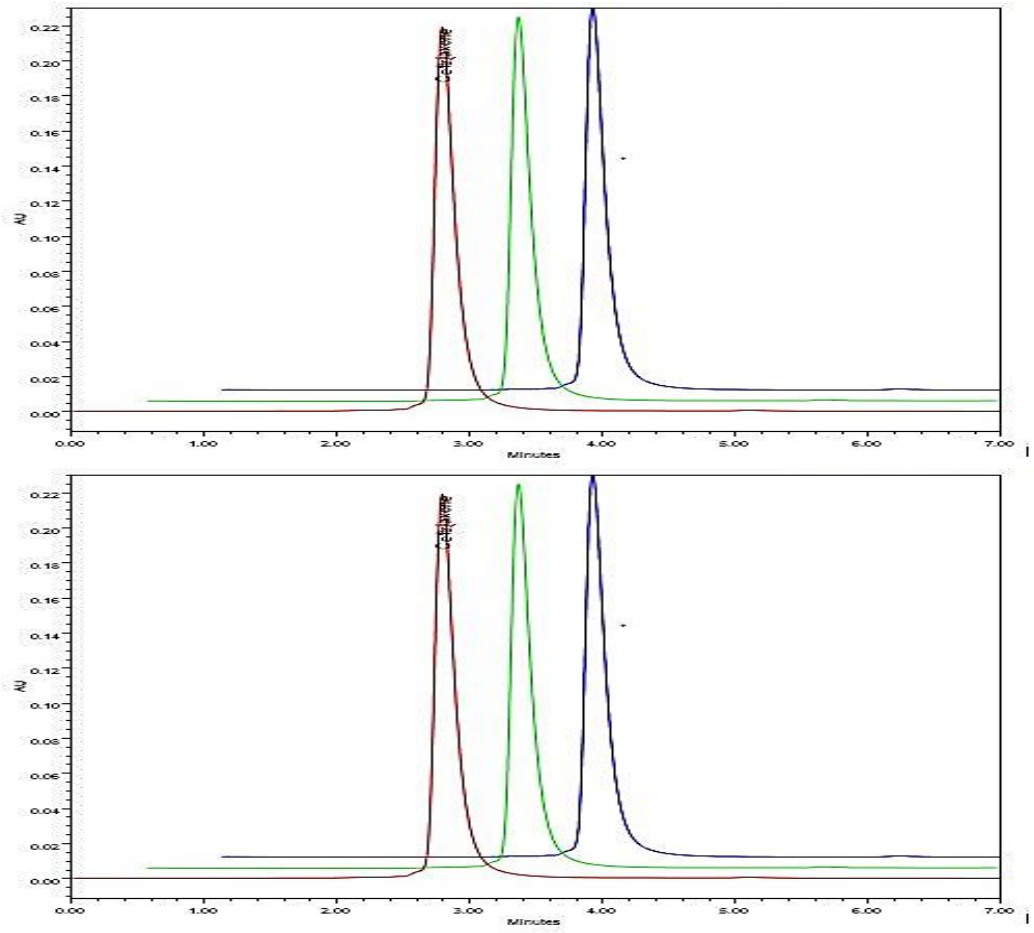

Figure 4: Chromatogram of (A) Cefxon injection; (B) Titan-1000 injection

Table 8: Accuracy and recovery calculation

Conc.	S-US	Mean of (S-US)	Added Amount	SD	% RSD	% Recovery	Mean % recovery
50% spiked	0.0194	0.0195	0.02	0.000014	0.072	99.16	99.19
	0.0195		mg/mL			99.21	
	0.0195					99.20	
100% spiked	0.0386	0.0387	0.04 mg/mL	0.000056	0.146	98.36	98.40
	0.0387					98.48	
	0.0386					98.35	
150% spiked	0.0589	0.0588	0.06 mg/mL	0.00016	0.282	98.97	98.84
	0.0589					98.91	
	0.0586					98.65	

Table 9: Calculation of Uncertainty

Parameter	%RSD	Square of %RSD
Linearity	1.041288	1.084281
Repeatability	0.5	0.25
Intermediate precision	0.22361	0.050
Practical Bias	0.039029537	0.001523
Sum of square		1.38580
Combined Uncertainty		1.177
Expanded Uncertainty		2.35

Conclusion

This new method is more cost-effective, complex, precise, and accurate while still being environmentally friendly. The results acquired during the verification process were accurate and within the ICH and pharmacopeia-provided system suitability parameters. The results were repeatable on multiple days and times. As a result of this, any laboratory or industry can use this developed method for ceftriaxone sodium in bulk or injection dosage form as long as they follow the method parameters and allowable variations as defined by the United States Pharmacopeia, British Pharmacopeia, and International Conference for Harmonization.

Table 10: Detailed summary of method validation parameters

Parameters	Obtained Results	Remarks
Linearity	Correlation coefficient (R^2) = (0.9999)	Method is Linear
Range	Correlation coefficient (R^2) = (0.9999)	0.01mg/mL to 0.08mg/mL
	%RSD = < 2	
Specificity	Chromatogram for the injection dosage form of Ceftriaxone Sodium was comparable and there was no interference by excipients of the dosage form.	Method is Specific
Precision	**%**RSD (Repeatability) = (0.5)	Method is Precise
	% RSD (Intermediate Precision) = (0.22361)	
Accuracy	%age Recovery was in the range of (98.403% to 99.191%)	Method is Accurate

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgments

The authors acknowledge the Higher Education Commission (HEC) of Pakistan for the support in the execution of this research.

References

1. Stoeckel K. Pharmacokinetics of rocephin®, a highly active new cephalosporin with an exceptionally long biological half-life. Chemother. 1981; 27(1):42-46.
2. Schaad UB, McCracken Jr GH, Loock CA, Thomas ML. Pharmacokinetics and bacteriologic efficacy of moxalactam, cefotaxime, cefoperazone, and rocephin in experimental bacterial meningitis. J Infect Dis. 1981; 143(2):156-163.
3. El-Maali NA, Ali AM, Ghandour MA. Electrochemical reduction and oxidation of two cephalosporin antibiotics: ceftriaxone (rocephin) and cefoperazone (cefobid). Electroanal. 1993; 5(7):599-604.
4. El-Maali NA, Ghandour M, Kauffmann J. Cephalosporin antibiotics at carbon paste and modified carbon paste electrodes in both aqueous and biological media. Bioelectrochem Bioenerg. 1995; 38(1):91-97.
5. Sar T, Mandal T, Samanta I, Rahaman A, Chakraborty A. Determination of ceftriaxone in plasma and ceftizoxime in milk of mastitic cows following single dose intravenous administration. Indian J Anim Sci. 2010; 80(12):1182-1184.
6. Hiremath B and Mruthyunjayaswamy B. Development and validation of a high-performance liquid chromatographic determination of ceftriaxone sodium and its application to drug quality control. Anal Lett. 2009; 42(14):2180-2191.
7. Richards D, Heel R, Brogden R, Speight T, Avery G. Ceftriaxone. Drugs. 1984; 27(6):469-527.
8. Yuk JH, Nightingale CH, Quintiliani R. Clinical pharmacokinetics of ceftriaxone. Clin Pharmacokin. 1989; 17(4):223-35.
9. Abdelaleem EA and Abdelwahab NS. Green chromatographic method for analysis of some anti-cough drugs and their toxic impurities with comparison to conventional methods. Saudi Pharm J. 2018; 26(8):1185-1191.
10. Ibrahim F, El-Deen AK, El Abass SA, Shimizu K. An ecofriendly green liquid chromatographic method for simultaneous determination of nicotinamide and clindamycin phosphate in pharmaceutical gel for acne treatment. J Food Drug Anal. 2017; 25(3):741-747.
11. Chakraborty I, Sathe SM, Khuman CN, Ghangrekar MM. Bioelectrochemically powered remediation of xenobiotic compounds and heavy metal toxicity using microbial fuel cell and microbial electrolysis cell. Mater Sci Energ Technol. 2020; 1;3:104-115.
12. Haq N, Alanazi FK, Alsarra IA, Shakeel F. Rapid Analysis of glibenclamide using an environmentally benign stability-indicating RP-HPLC method. Iranian J Pharm Res. 2014; 13(3):863.
13. Morelli B. Simultaneous determination of ceftriaxone and streptomycin in mixture by 'ratio-spectra'2nd derivative and 'zero-crossing'3rd derivative spectrophotometry. Talanta. 1994; 41(5):673-683.
14. Rahman M, Khan MA, Rub MA, Hoque MA. Effect of temperature and salts on the interaction of cetyltrimethylammonium bromide with ceftriaxone sodium trihydrate drug. J Mol Liq. 2016; 223:716-724.
15. Towfigh S, Pasternak J, Poirier A, Leister H, Babinchak T. A multicentre, open-label, randomised comparative study of tigecycline versus ceftriaxone sodium plus metronidazole for the treatment of hospitalised subjects with complicated intra-abdominal infections. Clin Microbiol Infect. 2010; 16(8):1274-1281.
16. Rahman M, Khan MA, Rub MA, Hoque MA. Effect of temperature and salts on the interaction of cetyltrimethylammonium bromide with ceftriaxone sodium trihydrate drug. J Mol Liq. 2016; 223:716-724.
17. Wen Q, Kong F, Zheng H, Yin J, Cao D, Ren Y, Wang G. Simultaneous processes of electricity generation and ceftriaxone sodium degradation in an air-cathode single chamber microbial fuel cell. J Power Sour. 2011; 196(5):2567-2572.
18. Zaki NM and Hafez MM. Enhanced antibacterial effect of ceftriaxone sodium-loaded chitosan nanoparticles against intracellular *Salmonella typhimurium*. J Am Assoc Pharm Sci. 2012; 13(2):411-421.
19. Farhadian N, Karimi M, Porozan S. Ceftriaxone sodium loaded onto polymer-lipid hybrid nanoparticles enhances antibacterial effect on gram-negative and gram-positive bacteria: Effects of lipid-polymer ratio on particles size, characteristics, *in vitro* drug release and antibacterial drug efficacy. J Drug Deliv Sci Technol. 2021; 63:102457.
20. Fey PD, Safranek TJ, Rupp ME, Dunne EF, Ribot E, Iwen PC, Bradford PA, Angulo FJ, Hinrichs SH. Ceftriaxone-resistant Salmonella infection acquired by a child from cattle. New Engl J Med. 2000; 342(17):1242-1249.
21. Chang YJ, Chen MC, Feng Y, Su LH, Li HC, Yang HP, Yu MJ, Chen CL, Chiu CH. Highly antimicrobial-resistant Nontyphoidal *Salmonella* from retail meats and clinical impact in children, Taiwan. Ped Neon. 2020; 61(4):432-438.

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

22. Jahan MS, Islam MJ, Begum R, Kayesh R, Rahman A. A study of method development, validation, and forced degradation for simultaneous quantification of paracetamol and ibuprofen in pharmaceutical dosage form by RP-HPLC method. Annal Chem Insgt. 2014; 9:75-81.
23. da Trindade MT, Kogawa AC, Salgado HRN. Turbidimetric Method: A multi-advantageous option for assessing the potency of ceftriaxone sodium in powder for injection. J AOAC Int. 2021; 104(1):204-210.
24. Trivedi HK, Kshtri N, Patel MC. A rapid, validated RP-HPLC method for the simultaneous determination of cleaning validation and cross-contamination of 12 beta-lactam compounds. Sci Pharm. 2013; 81(1):151-166.
25. Al-Zoman NZ, Maher HM, Al-Subaie A. Simultaneous determination of newly developed antiviral agents in pharmaceutical formulations by HPLC-DAD. Chem Central J. 2017; 11(1):1-8.