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RESEARCH ARTICLE

Spectrophotometric Determination and Cloud Point Extraction of Cefixime Drugs in Pure form and Pharmaceutical Preparation

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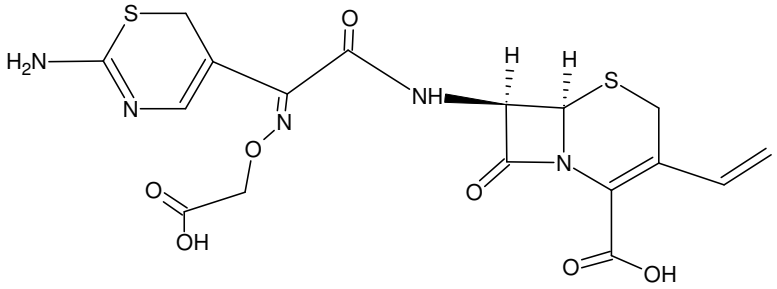
Abstract: A simple, rapid, accurate, sensitive and eco friendly method has been developed for the quantitative determination of cefixime(CFX) in pure form and pharmaceutical preparations by using a combination of cloud point extraction with UV-Visible absorption spectrophotometric method. Analytical applications of complexation with metal ions by reacting cefixime (CFX) with copper(II) and iron(III) to form chelate complexes under limited experimental conditions. The method based to dissolved CFX in 0.1 M NaOH, 10% (v/v) triton x-114 and mixed with (1000 μgmL^{-1}) copper(II) or (1000 μgmL^{-1}) iron(III). The formation of CFX-Cu(II) complex at pH 13 and wavelength 827 while the complex of CFX-Fe(III) was formatted at pH 11 and wavelength at 439 nm. The complexes of CFX-Cu(II) and CFX-Fe(III) obey Beer's Law in the range 10-130 and 10-160 µg/mL respectively. LOD and LOQ values for these complexes were 1.6906 µg/mL and 5.6355 µg/mL and LOQ values were 1.58655 µg/mL and 5.2887 µg/mL respectively. Method was validated and successfully applied to drug formulations like syrup infusion marketed in Amman and cefixime capsules marketed in Iraq. The results of analysis have been validated statistically by recovery studies and were found satisfactory.

Keywords: Antibiotic, Cefixime, Copper ion, Iron ion, Cloud point extraction

Introduction

Antibiotics are the chemotherapeutic agents that kill or inhibit the growth of microorganisms. This chemical agent is used to treat disease by destroying pathogenic microorganisms or inhibiting their growth at concentration low enough to avoid undesirable damage to the host. Antibiotics are drugs preparations which contain some chemical substances that are produced by microorganisms and by chemical synthesis. These substances at very low concentrations are known to totally destroy or partially inhibit microorganisms. Antibiotics have wide spread application in the treatment of bacterial disease[1] cefixime is the only oral third generation cephalosporin with a broad spectrum of antimicrobial effect on Haemophilus influenzae, *Moraxella catarrhalis*, *Neisseria gonorrheae*, *Escherichia coli* and *Klebsiella* resistant to ampicillin, other oral cephalosporins and trimethoprim-sulfamethoxazole. This characteristic

of cefixime permits its use in urinary and respiratory tract infections[2] cefixime (CFX)((6*R*,7*R*)-7-[(*Z*)-2-(2-amino-4-thiazolyl)-2-(carboxy-methoxyimino) acetamido]-8-oxo-3 vinyl-5-thia-1-azabicyclo-[4,2,0]-oct-2-ene-2-carboxylic acid), is a compound with potent mucolytic activity, for which it is used as an expectorant and broncho secretolytic in therapeutics[3]. The structures of drugs are shown in (Figure 1).



Formula ($C_{16}H_{15}N_5O_7S_2$), Mol.Wt, (453.452 g/mol)

Figure 1. The structure of cefixime[4]

It is third generation cephalosporin antibiotic. It is under the category of *ß*-lactam antibiotics/cell wall inhibitor. It acts by inhibiting an enzyme transpeptidase, involved in the building of bacterial cell walls. It is used in lower respiratory tract infections. It is helpful in acute urinary tract infections, biliary tract infections, sinusitis, acute otitis media, peptic ulcer and many more[4]. It is used to treat or prevent infections that are proven or strongly suspected to be caused by bacteria. One of the major problems with this drug is its very poor solubility in biological fluids that results into poor bioavailability after oral administration. It shows erratic dissolution problem in gastric and intestinal fluid due to its poor water solubility. Rate of absorption and/or extent of bioavailability for such insoluble drugs are controlled by rate of dissolution in gastrointestinal fluids[5] which describes a liquid chromatographic method for its assay in bulk form. In order to assure the quantity of cefixime in dosage forms, several methods have been reported which include liquid chromatography-mass spectrometry[6], high performance liquid chromatography[7-10], high performance thin layer chromatography[11,12], derivative spectrophotometry[13], voltammetry[14], and capillary electrophoresis[15]. The cloud point procedure (CPE) is based on the following phenomenon: an aqueous solution of some surfactant becomes turbid and separates into two isotropic phases if some condition such as temperature or pressure is changed or if an appropriate substance is added to the solution[16].

The aim of present work was to develop simple, economical, rapid, precise and accurate and eco friendly method for determination of CFX drug by using cloud point extraction.

Experimental

UV-Visible recording spectrophotometer SHIMADZU, Double beam UV-Vis, model UV-1800 made (Japan) with a response time of 0.1s was used for spectrophotomatric determination A quartz cell of 1 mL internal volume and 1 cm path length was used for absorbance measurements. Hotplate Stirrer (Hotplate stirrer Model L-81 Labincobv). Electric Balance (Sartorius, 4 digitals, made in Germany). OVEN (Memmert, maximum temperature 250, made in western Germany). Water Bath (A thermostat water Bath, model Unitemp) Centrifuge (Triup International corp, TRIU 800 Centrifuge, made in Korea). PH-meter (model BP 3001).

Drug and Materials

The chemicals used for this work are of high purity and used as received. Distilled water was used in the preparation of all solutions and for final rinsing of glass wares. A pure grade of cefixime was obtained from drug industries and Midical Appliance (SID) Samarra/ Iraq. A stock solution of 1000 μgmL^{-1} or ($2.205x10^{-3}$ M) for the drug cefix was prepared by dissolving 0.1 g in minimum amount of water and diluted to mark with water in a 100 mL volumetric flask. 0.1 M of NaOH (BDH, UK) was prepared from concentrated solution (1 M) by transferring10 mL into100 mL volumetric flask and diluted to mark with water. A stock solutions (1000 μgmL^{-1}) of copper ion(II)and iron ion(III) (95.5%, Sigma, USA) were prepared by dissolving 3.8 g of copper ion and 2.9 g of iron ion in 1000 mL volumetric class. Triton x-114 (purity >99.9%), was purchased from AMRESCO LLC (Solon, USA). A 10% (v/v) of Triton x-114 was prepared by diluting 10 mL with water in a 100 mL volumetric flask.

Recommended CPE procedure for cefix drug

Aliquots 10 mL of a solution containing known amount of cefixime drug was mixed with Cu^{+2} or Fe^{+3} ions. Then pH was adjusted by using 0.1 M NaOH and 10% (v/v) triton x-114. The mixture was shaken for 1 min and left to stand in a thermo-stated bath at 50 ^{o}C, for 20 min. Separation of the phases was achieved by centrifugation at 3000 rpm for 10 min, with stirring at 5 °C in ice bath the remaining of micellar phase was dissolved by ethanol, the measurements of absorbance of the complexes were followed by UV-Visible spectrophotometer with used 1.0 cm quartz cell at λ_{max} equal to 827 nm for CFX-Cu(II) complex and 439 nm for CFX-Fe(III) complex against blank which was prepared in the same way but without drug.

Preparation of pharmaceutical samples

Two types of pharmaceuticals for CFX namely capsules and syrup were obtained from the drugstores in Iraq and Amman. The powder of five capsules was mixed, homogenized and the content of one capsule (0.5339 g) which equivalent to 533.9 mg of active drug was dissolved in sufficient amount of water with continuous shaking and filtered. The filtrate solution was transferred into a 100 mL volumetric flask and diluted to mark with water. Solution contains 4000 μgmL^{-1} of CFX from which 1000 μgmL^{-1} was prepared by dilution. 25 mL containing different concentrations of the prepared sample solution were transferred to centrifugal tubes and each solution followed the recommended CPE procedure for cefix and the content of drug was measured spectrophotometrically at λ_{max} of 439 and 827 nm. The pharmaceuticals for syrup as each (5 mL) from drug contains (100 mg) cefixime. Solution is prepared by taking (5 mL) from syrup and dissolved in ethanol then solution is filtered and dilute in (100 mL) volumetric flask by distilled water, so that it gives (1000 μgmL^{-1}) from cefix. The same procedure is applied for syrup, CPE procedure for cefix and the content of drug was measured spectrophotometrically at λ_{max} of 439 nm and 827 nm.

Statistical analysis

Excel 2010 (Microsoft officer) was employed to carry out all statistical calculations.

Results and Discussion

Absorption spectra

In an attempt to ascertain the occurrence of reaction between two complexes in the reaction system, an absorption maximum at 827 nm (Figure 2) and 439 nm (Figure 3) which was

adopted of CPE for the drug. The absorption spectrum of the complex product formed was also recorded against the corresponding metal blank between 200 to 1100 nm before obtaining optimum conditions according to the recommended CPE procedure using a SHIMADZU, double beam UV-Vis, model UV-1800 with 1.0 cm quartz cell. It was observed that the absorption maximum of the colored product complex of cefix in 1.0 mL of 10% TX-114 occurred 827 nm, giving the molar absorptivities of $3x10^3$ $L.mol^{-1}.cm^{-1}$ for cefix drug with copper and $1.9x10^2 L.mol^{-1}.cm^{-1}$ for cefix drug with iron respectively. Thus the wavelength maximum at 827 nm and 439 nm for the cefix complex product was used throughout this study for ppm amounts.

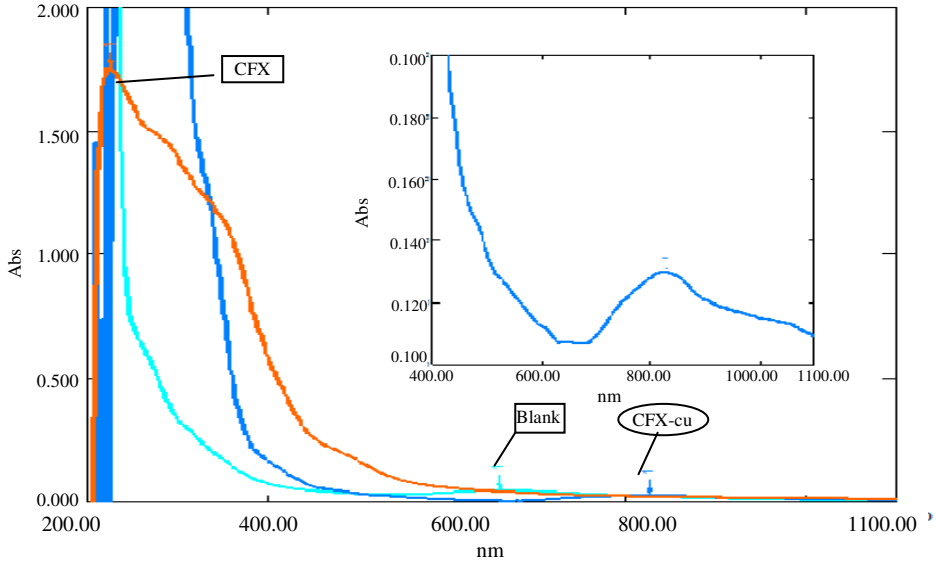


Figure 2. The absorption spectrum of the CFX-Cu(II) complex

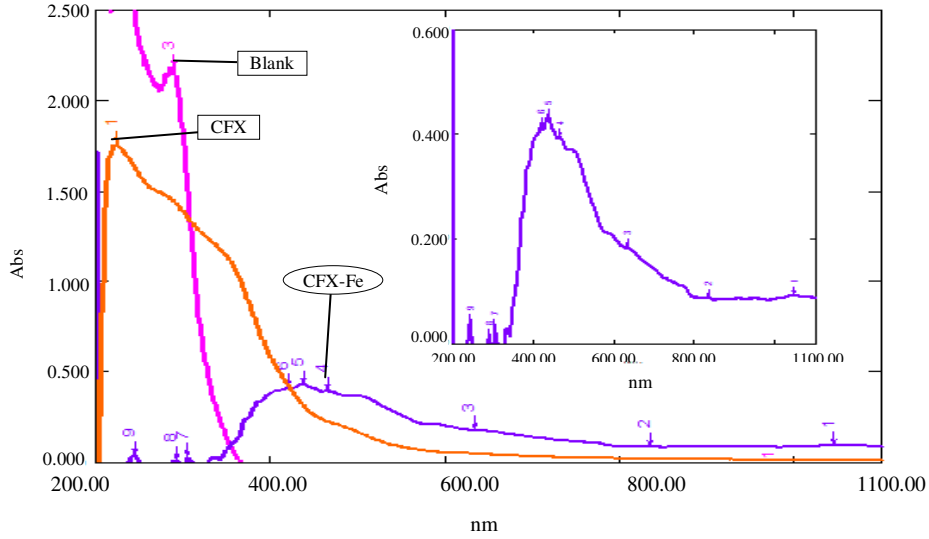


Figure 3. The absorption spectrum of the CFX-Fe(III) complex

Optimization of CPE methodology

A group of experiments has been conducted to study the effect of several variables that affect the extraction efficiency of the CPE and maximize the sensitivity of the detection system for drug under study using a classical optimization. The variables such as the concentration of metal ion, best of pH, best of buffer, best of volume buffer, triton x-114 amount, equilibration temperature and incubation time.

Effect of metal ions concentration

The effect of iron and cupric ion concentrations upon the absorbance values of the extracted complexes using 1000 µg/mL of drug solution. The optimum concentration of the metal ions that gave maximum absorbance was100 µg/mL of the optimum concentration of Cu(II) and Fe(III) ions were for complex. The absorbance is measured and the absorbance results are shown in Figure 4.

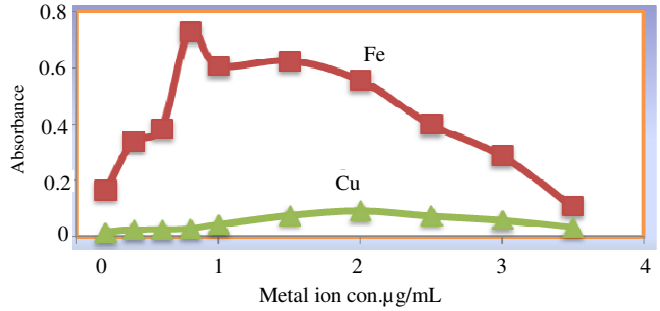


Figure 4. Effect of optimum concentration Cu(II) and Fe(III) ion concentration on absorbance of drug metal complexes

Effect of pH

The pH plays a unique role on metal ligand formation and subsequent extraction and is proved to be a main parameter for CPE[17], to find the best acidic function of the ion extraction process different value of pH 1-14. The results are shown in Figure 5, the best separation was achieved at pH =11 for Fe(III) and pH=13 for Cu(II). Show the value of absorbance intensity for the complexes drug- Cu and drug- Fe against the value of pH, the best values of pH recorded for the highest absorbance values were plotting of the absorbance values *versus* the value of pH is shown in Figure 5.

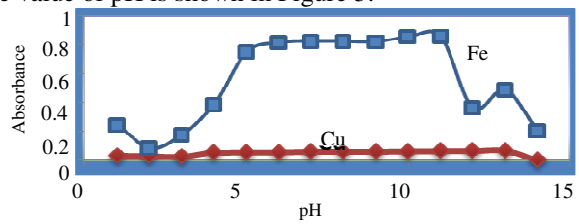


Figure 5. pH effect on the absorbance of drug- Cu(II) and drug- Fe(III) complexe

Effect of buffer solutions

The best values of buffer pH 13 recorded for the highest absorbance values were. The absorbance is measured the absorbance results are shown in Table 1 for complexes (Cu+ cefixime).

Table 1. Buffer pH 13

Preparation Buffer pH 13	Absorbance
Potassium buffer solutions	0.469

And the best values of buffer pH11 recorded for the highest absorbance values were the absorbance is measured. The absorbance results are shown in Table 2 for complexes (Fe+ cefixime).

Table 2. Buffer pH 11

Preparation buffer pH 11	Absorbance
Sodium *bicarbonate buffersolutions*	0.563
Sodium hydrogen ortho phosphate	0.268

Effect of volumes of buffer solutions

Figure 6 show the value of absorbance intensity for the complexes drug- Fe and drug-Cu against the value of buffer solutions, the best values of sodium bicarbonate buffer solutions recorded for the highest absorbance values, the best values of potassium buffer solutions recorded for the highest absorbance values.

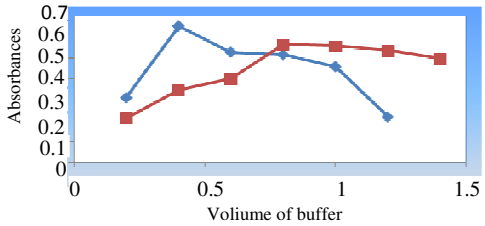


Figure 6. Buffer of pH effect on the absorbance of drug- Cu(II) and drug- Fe(III) complexe

Effect of type of surfactant with each metal cefixime

The type of surfactant plays very substantial role in cloud point extraction process where each surface owns spectral properties depend on practical basis of Micelles. Aliquots of 10 mL of a solution contains 1 mL cefixime, 2 mL Cu, 0.8 mL buffer pH 13 for copper metal and 1 mL cefixime, 0.8 mL Fe, 0.4 mL buffer pH 11 for Iron metal in 10 mL volumetric flask and used different surfactant for each drug (Tween 20, Tween 80, CTAP, SDS, triton x-100, triton x-114) at 50 ^{o}C for 20 min for cupric incubation time then it centrifugeted at 3000 rpm for 10 min, separated the surfactant- rich phase and dissolved in 1 mL ethanol then measured by UV-Vis at λ_{max}= 827 nm for Cu and 439 nm for Fe results shown in.

Table 3. Effect of surfactant type on absorbance

Absorbance at λ_{max} =439 for Fe(III)	0.293	0.539	0.476	0.428	0.344	0.743
Absorbance at λ_{max} =827 for Cu(II)	0.151	0.394	0.368	0.298	0.189	0.569

It was observed that triton x- 114 which have maximum absorbance at 439 nm is the best one for further study as shown in Table 3. Plotting the absorbance values of the cloud point *versus* the type of surfactant is shown in Figure 7 & 8.

Effect of triton x-114 amount

Most studies confirm that the amount of an non-ionic surfactant type TX-114 as an extracting medium plays an important role for maximizing the extraction efficiency by minimizing the

phase volume ratio (Vs/Va) and therefore improving the pre-concentration factor of the CPE procedure. Therefore, the amount of TX-114 was investigated by varying the volume of 10% TX-114 between (0.2-2.0 mL) for cefix. The results are presented in Figure 9. It was noticed that the absorbance values of cefix drug continued to increase dramatically and reached maximum at 1.6 mL of 10% TX-114 (*i.e.* 1.6% TX-114 in 10 mL solution) for Cu metal and1.0 mL of 10% TX-114 (*i.e.* 1. 0% TX-114 in 10 mL solution) for Fe metal. These values were selected as optimal amount and used in the proposed methods for the detection of cefix, plotting the absorbance values of the cloud point *versus* the volume of triton x-114 is shown in Figure 9.

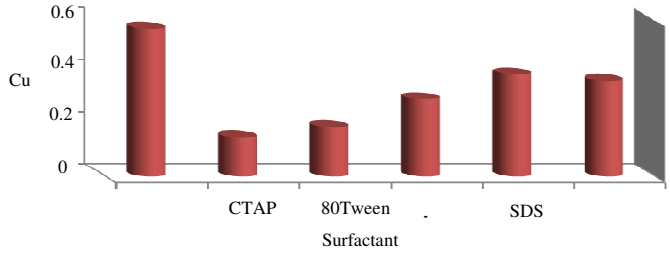


Figure 7. Type of surfactant for Cu(II)

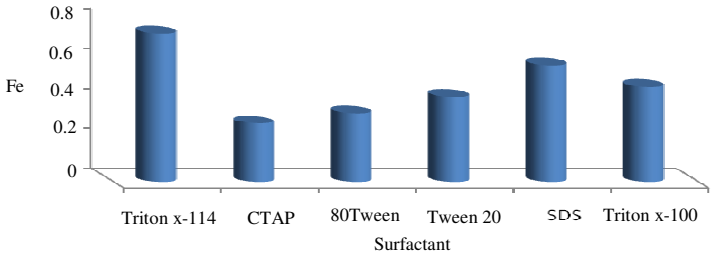


Figure 8. Type of surfactant for Fe(III)

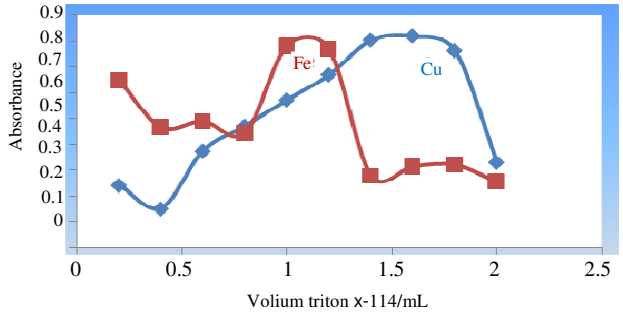


Figure 9. Effect of the TX-114 amount on absorbance of complexes product
(Conditions: for cefix: 100 µgmL^{-1}, metal Cu and metal Fe)

Effect of equilibration temperature and incubation time

The influence of these two parameters is considered of the most crucial steps in CPE, in order to ensure the efficient phase separation, which reflects certainly the magnitude of extraction efficiency of each target analyte. Figure 10 shows the variation on the absorption signal via varying the temperature between 35 to 80 ^{o}C at 20 min for incubation time for drug.

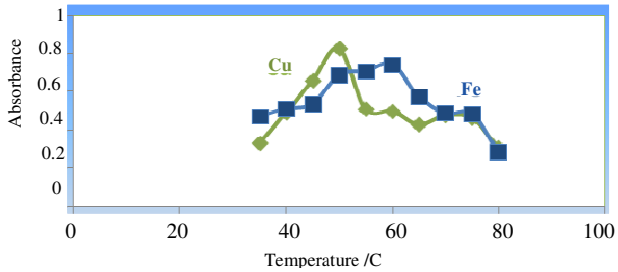


Figure 10. Absorbance *versus* temperature for Fe(III) and Cu(II)

The results show that the highest absorbency and extraction efficiency of the drug at temperature at 60 °C for cefixime with Fe(III), 50 °C for cefixime with Cu(II) for 20 min complexes then decreases in absorbance at higher temperature due to decomposition of product which reduces the extraction efficiency. This temperature is fixed in subsequent experiments.

Effect of the incubation time

Amount of 10 mL solution was prepared in volumetric flask containing, for Fe metal ion (1 mL cefixime, 0.8 mL Fe, 0.4 mL buffer pH 11 and 1 mL 10%(v/v) triton x-114) and for Cu metal (1 mL cefixime, 2 mL Cu, 0.8 mL buffer PH 13 and 1.6 mL 10%(v/v) triton x-114) then it was completed to the mark by distilled water, were mixed and the temperature was 60 °C for Fe and 50 °C for Cu and the incubation time varies from (5-50) min to form cloud point extraction then was measured by UV-Vis at λ_{max}= 439 nm for Fe and at λ_{max}= 827 nm for Cu Figure 11.

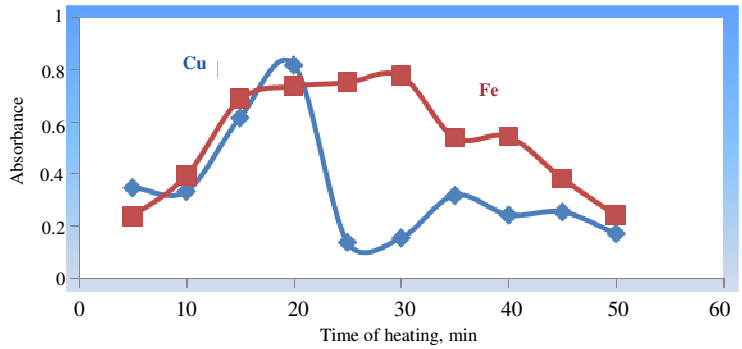


Figure 11. Absorbance *versus* time for Cu(II) and for Fe(III)

The time represents the amount of heat accumulated in the solution that allows Micelles lose water molecules in order to give small size hydrophobic with high viscosity easily entrap the product in it. It is clear that the optimum incubation time is 20 min for Cu respectively and maximum absorbance for all extracted Fe(III) complexes were observed after 30 min.

Order of additions

The effect of order for additions of the metal on the absorbance of each analyte by the general CPE was tested. Figure 12 shows that the best order of addition is the number 4 for target analytes due to giving a highest absorption signal among the others. The absorbance was measured and the absorbance results are shown in Table 4.

Table 4. Data of absorbance to order additions

No	Order additions	Absorbance at λ_{max} =439 for Fe(III)	Absorbance at λ_{max} =827 for Cu(II)
1	*D+ M+B+T*	0.767	0.824
2	*M+D+B+T*	0.684	0.174
3	*D+B+M+T*	0.607	0.087
4	*M+B+D+T*	0.732	0.788

Plotting of the absorbance values *versus* the order additions is shown in Figure 12.

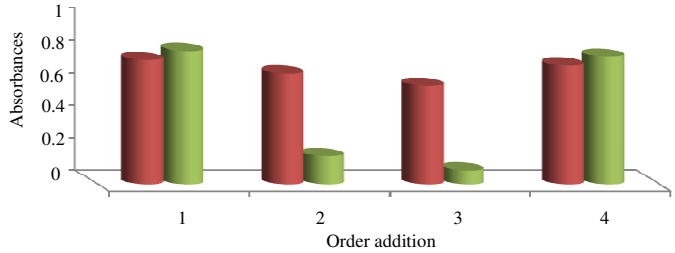


Figure 12. Effect of order additions for Fe (red) and Cu (green)

It is noted that the best addition is the first order of Fe(III) and the best addition is the four order of Cu(II), because if it's another order gets lost in the intensity of color and this order fixed in subsequent experiment.

Effect of organic solvents

Different organic solvents were examined to evaluate their effects on the intensity of the resulting complex and the data are shown in Table 5.

Table 5. Data of absorbance to solvents

No	Solvents	Absorbance at λ_{max} =439 for Fe	Absorbance at λ_{max} = 827 for Cu
1	Water	0.771	0.823
2	Ethanol	0.699	0.795
3	Methanol	0.623	0.363
4	Acetonitril	0.544	0.685
5	H_2O_2	0.377	0.210
6	chloroform	0.632	0.490
7	Acetyl aceton	0.105	0.203
8	Dimethyformamide	0.103	0.246
9	Dimethy phthalate	0.078	0.043
10	Dimethymalonate	0.016	0.021

Plotting of the absorbance values *versus* the solvent is shown in Figure 13.

It has been shown that water is the optimum solvent, economically, sensitivity method, cheap price, to provide and nontoxic. This solvent is fixed in subsequent experiment.

Effect of interference

The effect of some foreign organic compounds and inorganic compounds, which often found in environmental were studied by adding 1 mL of (100 ppm) equal amounts organic compounds, inorganic compounds to 1 mL of (100 ppm) of complex. The color was developed following the recommended procedure described earlier.

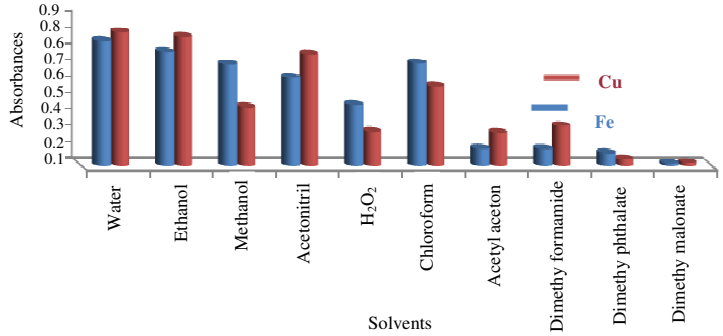


Figure 13. Effect of solvents for Fe and Cu

Table 6. Effect of interference

100 ppm Interference	Absorbance at λ_{max} =439 for Fe	Absorbance at λ_{max} = 827 for Cu
With out	0.772	0.822
Lactose	0.196	0.547
Starch	0.568	0.556
Arabic Gum	0.212	0.515
Talc	0.315	0.353
Glucose	0.248	0.385
$Ca_3(PO_4)_2$	0.201	0.202
$CaCO_3$	0.011	0.236

It was observed from Table 6 were not interfering with the determination at levels found in complex form.

Selected optimum conditions

After the study of the effect of different physical and chemical conditions on the absorbance intensity of the colored product, the optimum conditions for the proposed procedure were summarized in Table 7 and were used in all subsequent experiments.

Table 7. The optimum conditions for the determination of cefixime

Optimum	Concentrations	Range selected	Optimum quantities of complex (Cefxi-Fe)	Optimum quantities of complex (Cefxi- Cu)
λ_{max}, nm	-	190-1100	439	827
Effect of volume of metal ion required	1000 ppm	0.2 -3.5 mL	0.8 mL	2 mL
Effect of PH	0.1 M (NaoH)	1-14	11	13
Buffer pH	-	-	Sodium bicarbonate buffer solutions	Potassium buffer solutions
Effect of volume of Buffer	-	0.2-1.6 mL	0.6 mL	0.8 mL
Effect of volume of triton x114 required	10%(v/v)	0.2 -2.0 mL	1 mL	1.6 mL
Effect of time heating	-	5-60 min	30 min	20 min
Cefixime solution required	1000 ppm	10 -160 ppm	1mL	1 mL

Preparation of calibration curve in CPE

Amount of 10 mL solution was prepared containing increasing concentration of drug cefixime by taking (10-160) μgmL^{-1} cefixime, 0.8 mL Fe, 0.4 mL buffer pH 11 and 1 mL 10% (v/v) triton x-114 and for Cu metal 2 mL was taken 0.8 mL buffer pH 13, 10-130 μgmL^{-1} cefixime and 1.6 mL 10%(v/v) triton x-114 then it was completed to the mark by distilled water and mixed, heated at optimum temperature in the thermostat water bath at optimum incubation time, to form cloud point then aqueous phase was separated by centrifugation at 4000 rpm for 20 min, 1 mL ethanol was added to the surfactant-rich phase to dissolve it then is measured by UV-Vis at λ_{max}= 439 nm for iron and at λ_{max}= 827 nm for cupric, triplicate manner. The absorbance measurements are illustrated in Table 8 and Table 9

Table 8. The absorbance measurements of standard solutions of complex (CFX-cu)

Conc. ppm	Mean Absorbance	RSD%	Found	Recovery,%
10	0.155	1.2903	13.1184	131
20	0.219	2.7775	21.5394	107
30	0.286	0.3496	30.3553	101
40	0.345	0.7668	38.1184	95
50	0.417	0.4796	47.5921	95
60	0.497	0.2012	58.1184	96
70	0.582	0.4545	69.3026	99
80	0.661	0.1512	79.6973	99
90	0.742	0.2695	90.355	100
100	0.823	0.7390	101.013	101
110	0.897	0.2949	110.73	100
120	0.986	0.8049	122.46	102
130	1.034	0.5117	128.77	99

Table 9. The absorbance measurements of standard solutions of complex (CFX-Fe)

Conc. ppm	Mean Absorbance	RSD%	Found	Recovery,%
10	0.085	3.1126	9.8732	98
20	0.152	1.3157	19.3098	96
30	0.21	1.7169	27.478	91
40	0.301	0.3322	40.2957	100
50	0.39	0.4441	52.8309	105
60	0.439	1.2053	59.7323	99
70	0.497	0.2012	67.9014	97
80	0.589	0.4491	80.859	101
90	0.671	0.2980	92.4081	102
100	0.732	0.3614	101	101
120	0.845	0.5158	116.915	97
140	1.011	0.0989	140.295	100
160	1.143	0.0874	158.88	99

The calibration curve was plotting the mean absorbance values of the cloud point *versus* the concentration (ppm) of CFX- cupric as shown in Figure 14.

The calibration curve was plotting the mean absorbance values of the cloud point *versus* the concentration (ppm) of CFX- Iron as shown in Figure 15.

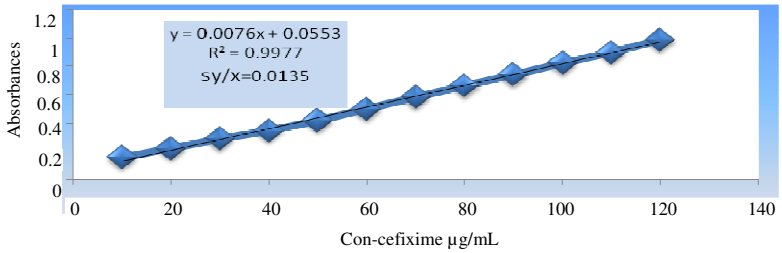


Figure 14. (Cefixime + Cu) calibration curve

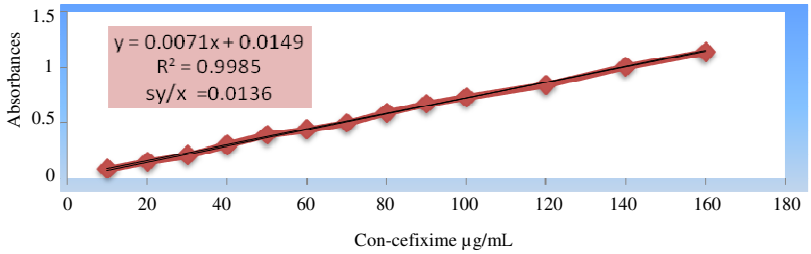


Figure 15. (Cefixime + Fe) calibration curve

Optical characteristics features of the calibration curve

Table 10 shows the main features of the calibration curve and measuring the absorbance at 827 nm and 439 nm.

Table 10. Optical characteristic features of calibration curve

Parameter	Complex (cefixime-Fe)	Complex (cefixime-Cu)
Color of product	orange	Green
Wave length λ_{max}, nm	439	827
Concentration rang, µgmL^{-1}	(10-160 µgmL^{-1})	(10-130 µgmL^{-1})
Regression equation	y=0.0071x +0.0149	y =0.0076x +0.0553
Correlation coefficient(r)	0.9992	0.9988
Correlation coefficient (r^2)	0.9985	0.9977
Variation coefficient, %	99.85	99.77
Limit of detection, µgmL^{-1}	1.5865	1.6906
Limit of quantitation, µgmL^{-1}	5.2887	5.6355
Sandell's sensitivity, µg cm^{-2}	0.2320	0.1315
Slope (m)	0.0071	0.0076
Intercept (C)	0.0149	0.0553
Molar absorptivity, L.mol^{-1}.cm^{-1}	1.9×10^{2}	3×10^{3}
Composition of product	1:1	1:1
C.L for slope (b±tSb) at 95%	$0.0071\pm1.8682\times10^{-4}$	$0.0076\pm2.42\times10^{-3}$
C.L for intercept (a±tSa) at95%	0.0149±0.016229	0.0553±1.74739
C.L for conc.30 µgmL^{-1} at 95%	$27.478\pm2.8\times10^{-6}$	$30.3553\pm2.48\times10^{-6}$
C.L for conc.60 µgmL^{-1} at 95%	$59.7323\pm4.1\times10^{-6}$	$58.1184\pm2.48\times10^{-6}$
C.L for conc.90 µgmL^{-1} at 95%	$92.4081\pm4.9\times10^{-6}$	$90.355\pm4.96\times10^{-6}$
C.L for conc.120 µgmL^{-1} at 95%	$116.915\pm3.4\times10^{-5}$	$122.46\pm6.20\times10^{-5}$

Stoichiometric determination of color complex

Continuous variation method (*Job`s method)*

A series of 1, 2, 3, 4, 5, 6, 7, 8 and 9 mL of 1×10^{-4} mol L^{-1} of the solution that contain cefixime was ipette into each of 10 mL volumetric flask then 9, 8, 7, 6, 5, 4, 3, 2 and 1 mL of 1×10^{-4} mol $L^{-1}$of metal the absorbance of the solution was measured by UV-Vis spectrophotometer at λ_{max} 827 nm and 439 nm the stoichiometric ratio between cefixime with metal 1:1 results are shown in the Table 11.

Table 11. The continuous variation method of cefixime with metal (Copper) complex

V D mL	V M mL	VD / VT	Absorbance at λ= 827 for color compound	Absorbance at λ= 439 for color compound
1	9	0.1	0.142	0.067
2	8	0.2	0.313	0.159
3	7	0.3	0.431	0.290
4	6	0.4	0.549	0.357
5	5	0.5	0.643	0.519
6	4	0.6	0.52	0.479
7	3	0.7	0.356	0.278
8	2	0.8	0.121	0.140
9	1	0.9	0.034	0.032

Plotting the value of absorbance *versus* the VD/VT is shown in Figure 16

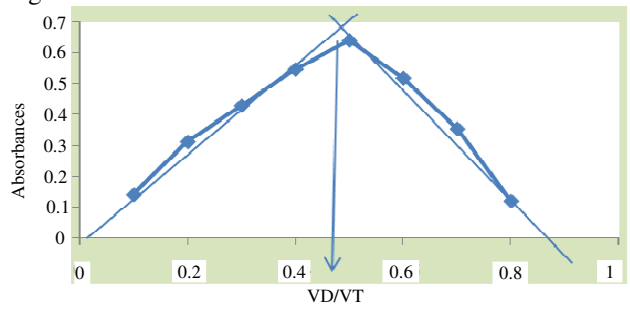


Figure 16. Continuous variation method plot

VD: values of the compound (Cefixime), V M: The values of the metal (Copper). VT: Total (V M+V D)

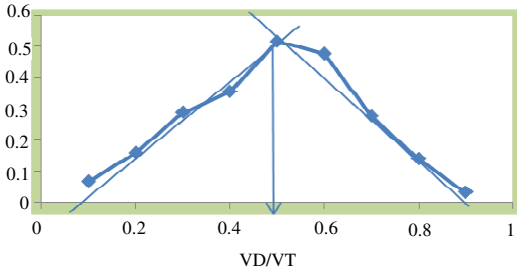


Figure 17: Continuous variation method plot

VD: values of the compound (Cefixime), V M: The values of the metal (Iron). VT: Total (V M+V D)

Mole -Ratio method

Aliquots of 10 mL solution containing 1×10^{-4} mol L^{-1} of 1 mL cefixime and increasing concentrations 1×10^{-4} mol L^{-1} of 0.2,0.4, 0.6, 0.8, 1.0, 1.2, 1.4, 1.6 and 1.8 mL of Cu and Fe 2×10^{-6}-2×10^{-5} mol L^{-1} metal. The absorbance of the solutions were measured by UV-Vis spectrophotometer *versus* blank at λ_{max}=827 and 439 nm the stoichiometric ratio between 1:1 results are shown in the Table 12.

Table 12. The mole-ratio method of the cefixime with copper and iron

CL	CL/CM	Absorbance at λ= 827 nm	Absorbance at λ= 439 nm for color compound
2×10^{-6}	0.2	0.032	0.098
4×10^{-6}	0.4	0.094	0.149
6×10^{-6}	0.6	0.121	0.192
8×10^{-6}	0.8	0.153	0.231
1×10^{-5}	1.0	0.201	0.248
1.2×10^{-5}	1.2	0.194	0.241
1.4×10^{-5}	1.4	0.199	0.235
1.6×10^{-5}	1.6	0.195	0.229
1.8×10^{-5}	1.8	0.199	0.218

Plotting the value of absorbance *versus* the CL/CM is shown in Figure 18.

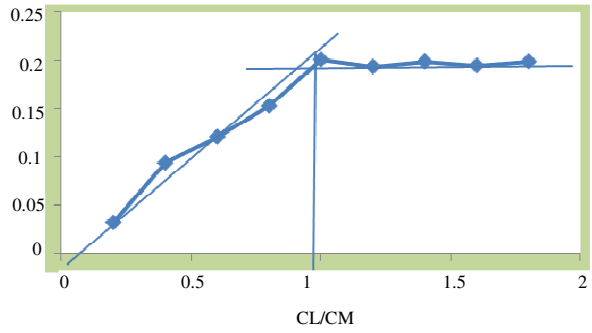


Figure 18. Mole - Ratio plot of cefixime and copper complex

CL: concentration of the metal (Copper), CM: concentration of the compound (Cefixime)

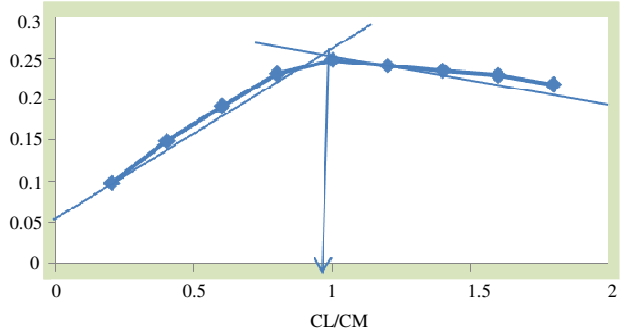


Figure 19. Mole-ratio plot of cefixime and iron complex

CL: concentration of the metal (Iron), CM: concentration of the compound (Cefixime)

Applications of the cloud point extraction on pharmaceuticals

CPE has been applied on pharmaceutical cefixime, the manufacture company (Novartis) that contains (533.9 mg) from cefixime. The results are good and of high reliability in the analysis of samples in the pharmaceutical preparation. The results are summarized in the Table 13 for cefixi.

Table 13. Data for determination cefix with cupric in the pharmaceutical preparation capsule (cefixime) by CPE

Amount of cefix/µg mL^{-1}	Mean absorbance	Relative stander deviation (RSD)	*Found	Recovery %	Average Recovery%	Erel %	Average Erel%
30	0.263	0.3802	27.32	91.0		-8.9	
60	0.487	0.4106	56.80	94.6	90.8	-5.3	-9.03
90	0.651	0.1536	78.38	87.0		-12.9	
[*]= Average of three							

The proposed method is also applied on syrup cefixime the manufacture company is Novartis. As each (5 mL) from drug contains (100 mg) cefixime, we get good and high reliability results that are summarized in the Tables 14-16 for cefix by CPE.

Table 14. Data for determination cefix with cupric in the pharmaceutical preparation syrup (cefixime) by CPE

Amount of cefix/ µgmL^{-1}	Mean absorbance	Relative stander deviation (RSD)	*Found	Recovery%	Average Recovery%	Erel%	Average Erel%
30	0.270	0.3703	28.25	94.1		-5.8	
60	0.479	0.3615	55.75	92.9	93.8	-7.0	-6.06
90	0.702	0.3768	85.09	94.5		-5.4	

Table 15. Data for determination cefix with Iron in the pharmaceutical preparation capsule (cefixime) by CPE

Amount of cefix/ µgmL^{-1}	Mean absorbance	Relative stander deviation (RSD)	*Found	Recovery%	Average recovery%	Erel%	Average erel%
30	0.215	1.2305	28.18	93.9	97.6	-0.6	
60	0.430	0.2325	58.46	97.4		-2.5	-6.8
90	0.665	0.2604	91.56	101.7		1.7	

Table 16. Data for determination cefix with iron in the pharmaceutical preparation syrup (cefixime) by CPE

Amount of cefix/ µg mL^{-1}	Mean absorbance	Relative stander deviation (RSD)	*Found	Recovery%	Average recovery%	Erel%	Average erel%
30	0.210	0.9523	27.47	91.5	94.0	-8.4	
60	0.394	0.2538	53.39	88.9		-11.0	-5.1
90	0.680	0.3890	93.67	104.0		4.0	

Conclusion

The proposed method is simple, sensitive and free from drastic experimental conditions such as heating. It is also accurate, precise enough to be successfully adopted as an alternative to the existing spectrophotometric method and evaluation of cefixime in a metal Using CPE and in pharmaceutical Preparation samples.

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