

Comparison of Three Independent Methods of Cephalexin Determination by Means of Potassium Caroate

Comparación de tres métodos independientes para la detección de cefalexina usando el potasio de caroata

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ABSTRACT

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Aims. Three different assays for the quantity determination of Cephalexin in pure substance were developed and compared using potassium caroate as analytical reagent.

Materials and Methods. Cephalexin pure substance was used and analytical reaction by means of KHSO_5 as an oxidant was studied. Voltammetric, kinetic-spectrophotometric and iodometric methods were developed and validated.

Results. The chemical reaction is finished with formation of the S-oxide of Cephalexin in the iodometric and voltammetric methods and the S-oxide hydrolysis product in the kinetic-spectrophotometric method. The reaction finishes in 1 min (observation time). The methods have been validated and good results including precision, accuracy and recovery were obtained. The recovery percent ranged from 98.7 to 101.4%, RSD from 1.42 to 3.0%. The methods proposed are linear in a wide range: voltammetry 2-45 $\mu\text{g mL}^{-1}$, kinetic-spectrophotometry 1-16 $\mu\text{g mL}^{-1}$, and iodometry 0.05-0.35 mg mL^{-1} . LOD and LOQ were calculated. The kinetic-spectrophotometric method is the most sensitive (LOQ=1.0 $\mu\text{g mL}^{-1}$).

Conclusions. The methods proposed are rapid, simple and inexpensive and could be applied to pharmaceutical preparation.

Key Words: Cephalexin, iodometry, kinetic-spectrophotometry, potassium caroate, voltammetry.

RESUMEN

Objetivo. Se han desarrollado tres métodos diferentes para la detección cuantitativa de Cefalexina en producto puro, usando el caroata de potasio como un reactivo analítico.

Materiales y métodos. Fue estudiada la reacción analítica de cefalexina con KHSO_5 como oxidante. Se han desarrollado y validado los métodos de voltamperimetría, de espectrofotometría cinética y de iodometría.

Resultados. La reacción química se completa con la formación de s-óxido de Cefalexina dentro de los métodos de iodometría y de voltamperometría, y con la formación del producto de hidrólisis de s-óxido en el método de espectrofotometría cinética. La reacción se completa en 1 minuto (tiempo de observación). Los métodos han sido validados, y se han obtenido resultados buenos, incluida la exactitud, la fiabilidad y la reproducibilidad. El contenido de la sustancia básica oscila desde 98,7% hasta 101,4%. El RSD está entre 1,42 y 3,0%. Los métodos propuestos son lineales en un amplio espectro: la voltamperometría 2-45 $\mu\text{g mL}^{-1}$, la espectrofotometría cinética 1-16 $\mu\text{g mL}^{-1}$, y la iodometría 0,05-0,35 mg mL^{-1} . Han sido calculados el límite de detección y el límite de cuantificación. El método de espectrofotometría cinética es el más sensible (el límite de cuantificación = 1,0 mcg mL^{-1}).

Conclusiones. Los métodos propuestos son rápidos, simples y baratos, y pueden ser utilizados en las preparaciones farmacéuticas.

Palabra clave: Cefalexina, iodometría, voltamperometría, caroata de potasio, espectrofotometría cinética



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INTRODUCTION

Cephalexin monohydrate is a derivate of aminodesacetoxyccephalosporane acid, belongs to β -lactamic antibiotics of the I generation with a range of pharmacological activity. Cephalexin has a great Gram-negative and Gram-positive, antimicrobial properties. It is produced in gelatin capsules and in the form of suspension. Cephalexin was included into the "Top 200 Drugs" mostly prescribed in the USA according to National Data Corporation Health¹.

The British Pharmacopeia and the State Pharmacopeia of Ukraine recommend a potentiometric nonaqueous method for the determination of Cephalexin-standard using perchloric acid as the titrant and HPLC method using a mixture of phosphate buffer, methanol and acetonitrile as mobile phase with UV detection at 254 nm^{2,3}.

Several procedures have been reported in the literature for the analysis of Cephalexin. The reported methods include UV-spectroscopic methods and visible spectroscopic methods^{4,5}, spectrofluorimetry^{6,7}, chemiluminescence⁸, atomic absorption spectrometric methods⁹, polarography¹⁰⁻¹². The impressive increase in the use of HPLC in the past thirty years did not pass β -lactam antibiotics. HPLC has been used often in all fields of β -lactam research, not only as an assay method but also as a tool for purification of the antibiotics. Due to the insolubility of these compounds in organic solvents, normal phase LC was sparingly used^{13,14}. But, HPLC methods are quiet expensive and product a big amount of wastes including toxic organic solvents¹⁵. Some of these methods involve several manipulation steps, which are not simple for routine analysis of pharmaceutical formulations and need advanced instruments.

The direct spectrophotometric methods suffer a lack of specificity because all compounds containing the β -lactam ring absorb in the range 250-270 nm.

The official rules in pharmaceutical preparations use iodometric titration are unselective as all antibiotics, the first substances for their production as well as their degradation products are being oxidized by iodine. It is not suitable for control of the purity of the antibiotics of this group¹⁶. The peculiarity of this method is the dependence of the titer on the conditions.

Therefore, the development of precise, correct and low cost technique for drugs analysis is of a great interest. Titrimetric, kinetic-spectrophotometric method and voltammetry may serve as useful alternative to many of the previously mentioned new sensitivity, remarkable accuracy and precision and wide applicability.

The present work aim is to develop simple, rapid and cost-effective methods for the determination of Cephalexin in pure form and dosage forms using titrimetric, kinetic-spectrophotometric and voltammetric techniques. The methods used KHSO_5 as a reagent. Moreover, the developed methods meet the requirements of the concept of Green Chemistry.

MATERIALS AND METHODS

All materials were of the analytical reagent grade, and the solutions were prepared with twice-distilled water. Cephalexin pure substance "Purilex" ((6R, 7R)-7-[(R)-2-amino-2-phenylacetamido]-3-methyl-8-oxo-5-tia-1-azabicyclo[4.2.0]oct-2-en-2-carbonic acid monohydrate) produced by DMS Anti-infectives Chemferm S.A., Spain: ser. B425055; assay 100.9 % (BPh); with 6.2 % content of water). Granules for suspension preparation for internal application "Cephalexin" 250 mg/5 mL, "Hemopharm" AD, Serbia, series 3130408, content: 5 mL of suspension contain 250 mg of cephalexin monohydrate, excipients. The acceptable range is 225-300 mg/5 mL.

The oxidant was KHSO_5 , potassium caroate in the form of a triple potassium salt of Caro's acid, $2\text{KHSO}_5 \cdot \text{KHSO}_4 \cdot \text{K}_2\text{SO}_4$ (Acros Organics). The choice of the reagent was determined by its rather high oxidative capacity, $E^0 = 1.84$ V, easy availability, and satisfactory solubility in water, and also by sufficiently high stability in the use and storage.

Preparation of standard solutions

Preparation of 0.02 M potassium caroate solution. 0.615 g of $2\text{KHSO}_5 \cdot \text{KHSO}_4 \cdot \text{K}_2\text{SO}_4$ was dissolved in twice-distilled water (100 mL). The concentration of potassium caroate was controlled by iodometric titration.

Preparation of 0.01 M sodium hydroxide solution. 0.4 g of sodium hydroxide was weighted and dissolved in 100 mL of distilled water. A 1/10 dilution was made to obtain required concentration.

Preparation of 0.02 M sodium thiosulfate solution. A 0.1 M solution of sodium thiosulfate was prepared from the standard titre fixanal. A 2/10 dilution was made to obtain required concentration.

Preparation of 5 % potassium iodine solution. 5.0 g of sodium hydroxide was weighted and dissolved in 100 mL of distilled water.

Preparation of 0.1 M sulfuric acid solution. The solution was prepared from the standard titre fixanal in a 500 mL volumetric flask.

Polarograms were recorded in a 0.03 M supporting acetate buffer solution with pH 4.5. To prepare it, 23.0 g of $\text{CH}_3\text{COONa} \cdot 3\text{H}_2\text{O}$ of chemically pure grade were dissolved in 50 mL of twice-distilled water, and pH 4.5 was set using 0.1 M HCl; the mixture was diluted to 500 mL with twice-distilled water and stirred carefully.

Preparation of cephalexin working standard solutions

Seven solutions of the following concentrations: 80%, 85%, 90%, 95%, 100%, 110%, 120% were prepared. A precisely weighed portion (0.2388 g; 0.2538 g; 0.2687 g; 0.2836 g; 0.2985 g; 0.3284 g; 0.3582 g correspondingly) of the substance was transferred into a 100-mL volumetric flask and diluted to the mark.

Apparatus

Spectrophotometry. A spectrophotometer SF-46 (LOMO, JSC, St-Petersburg, Russia). 1 cm quartz cells were used for spectral measurements.

Voltammetry. Voltammograms were recorded on a CLA-3 oscillographic polarography (Rostov-on-Don Research Institute, Russia) in a three-electrode thermostated cell at 20°C; the indicator electrode was dropping mercury one, the reference electrode was a saturated calomel electrode, and the auxiliary electrode was made of platinum. The potentials of peak maxima were measured with V7-21 digital voltmeter with the precision ± 1 mV. Triangular-shaped polarizing voltage was applied on the cell electrodes at a scanning rate of $v=0.5$ V/s. The potential was varied in the range from -0.2 to -0.4 V. The dissolved oxygen was removed from solutions by blowing purified argon over 20 min. pH values were measured with an ESL-43-07 glass electrode against an EVL-1M3.1 silver-silver chloride reference electrode using an I-160 M laboratory potentiometer (Belarus').

Titration. The titrant volume was measured using a 10 mL microburette with the accuracy of ± 0.01 mL.

Procedure

Kinetic-spectrophotometric method. 0.35 g portion of the preparation powder was dissolved in twice-distilled water (100 mL). A 10.00 mL aliquot of the solution obtained, 2.0 mL of 0.02 M potassium caroate solution and 2.0 mL of 0.01 M of sodium hydroxide was transferred into 100 mL volumetric flask. The volume was brought to the mark and mixed. After the addition of NaOH a stop watch was switched on. Absorbance measurements at 305 nm were carried out every 2 min, during a total of 15 min after the preparation of the samples.

Voltammetric method. 0.5000 g portion of cephalexin substance was dissolved in twice-distilled water (100 mL). A 5.5 mL portion of a $2.0 \cdot 10^{-2}$ M of potassium caroate solution was added to 10 mL of the solution prepared, and the mixture was stirred carefully. A 5.0 mL portion of the solution prepared was transferred into a 50 mL volumetric flask, 5.0 mL of a 0.3 M acetate buffer solution with pH 4.0 was added, and the mixture was diluted to the mark with twice-distilled water and stirred carefully. The solution was charged into an electrolyser and voltammogram was recorded in the range from -0.2 to -1.4 V.

Iodometric method. 0.36 g of cephalexin substance was dissolved in a twice-distilled water (1000 mL). A 10.00 mL portion was transferred into 100 mL volumetric flask, 10.00 mL of 0.02 M KHSO_5 solution was added and brought to the mark, stirred vigorously and left for 1 min. A 10.00 mL aliquot of the mixture obtained was transferred using the pipette into titration flask, 1 mL of 0.1 M sulfuric acid solution and 1 mL of 5 % potassium iodine solution were added. The isolated iodine was titrated by 0.02 M the titrated solution of sodium tiosulfate (V , mL). The control experiment was carried out in the same conditions paralleled (without Cephalexin with the same amount of KHSO_5 0.02 M solution (V_0 , mL). Each one mL of 0.01 M solution of sodium tiosulphate is equivalent to 0.003654 g of cephalexin monohydrate ($\text{C}_{16}\text{H}_{17}\text{N}_3\text{O}_4\text{S}$) (content of cephalexin dehydrate in the limits 95-101%).

Method validation

The method was validated according to the guidelines of the International Conference on Harmonization¹⁷.

The Precision and Accuracy on these procedures were investigated with respect to repeatability and determined by performing five repeated analysis of the samples on the same day, under the same experimental conditions.

The Linearity was determined for a wide range of concentration for each of the methods. The calibration curves for each method were obtained, each research comprises 7 experimental points.

LOD and LOQ were calculated from regression equation as $3.3 S_0/b$ and $10 S_0/b$ respectively, where S_0 and b are standard deviation slopes of the calibration curve.

Method comparison. Results obtained in this study were compared to those given in the quality certificates for the pure substance, i.e. HPLC method. The F-data comparison was performed between repeatability of the results obtained by iodometric method and by proposed kinetic-spectropho-

tomteric and voltammetric methods. The F-values should be less than 6.39.

RESULTS AND DISCUSSION

Kinetic-spectrophotometric method

In a basic medium Cephalexin S-oxide undergoes hydrolytic cleavage. Figure 1 shows the electronic spectra of Cephalexin and the reaction product. The appearance of a new band with absorption $\lambda_{\max} = 305$ nm demonstrates its formation in the reaction of alkaline hydrolysis of Cephalexin S-oxide in the presence of potassium caroate (perhydrolysis reaction).

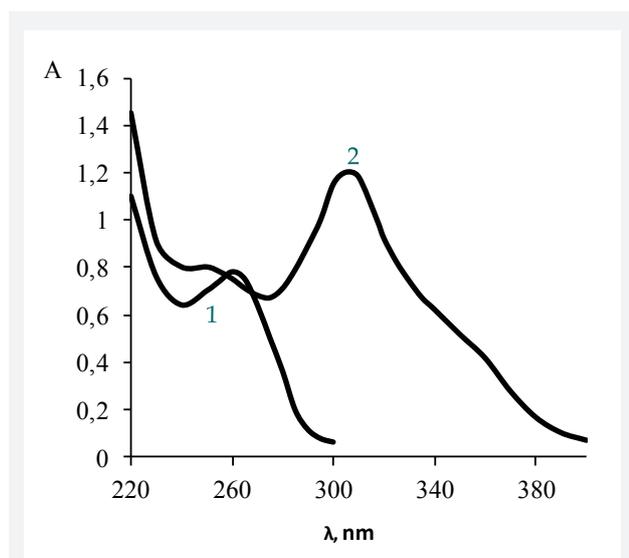


Fig. 1. Electronic spectra of Cephalexin (1) and S-reaction and perhydrolysis conjugated reaction with potassium caroate (2) absorbance.

$c(\text{CF}) = 1 \times 10^{-4} \text{ M}$; $c(\text{KHSO}_5) = 2 \times 10^{-4} \text{ M}$; $c(\text{NaOH}) = 0.01 \text{ M}$

The interaction between Cephalexin and potassium caroate in acidic medium ($\text{pH} = 3-5$) is stoichiometric and fast: 1 mol of Cephalexin per 1 mol of KHSO_5 (observation time is 1 min). In the basic medium a perhydrolysis conjugated reaction proceeds with formation of a corresponding product (Fig. 2, b).

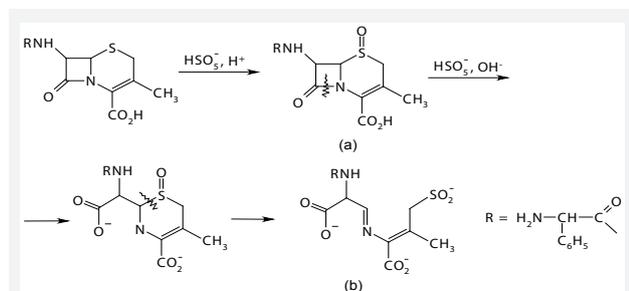


Fig. 2. The scheme of Cephalexin transformation: a) S-oxide product, b) S-reaction and perhydrolysis conjugated reaction with potassium caroate

During the experiment, it was determined that the order of mixing influences the kinetics and yield of the reaction. The highest rate of product accumulation was observed only after a prior addition of a potassium caroate solution to cephalosporin solution (formation of Cephalexin S-oxide), and then to a basic solution, optimal concentrations: potassium caroate $8 \cdot 10^{-4} \text{ M}$, alkaline $2.1 \cdot 10^{-2} \text{ M}$ at 298-299 K. The reaction product wasn't formed without oxidant during the first 30 min. The product formation rate was assessed by the slope ($\text{tg}\alpha, \text{min}^{-1}$) of linear plots of kinetic absorption curves, A versus time, t (in minutes).

Validation of the proposed method has been carried out for the parameters like linearity, precision, accuracy, LOD and LOQ. Detection wavelength selected for analysis was 305 nm. Linearity has been studied over a small drug concentration range from $1-50 \mu\text{g mL}^{-1}$. The correlation coefficient $r=0.999$ obtained for regression line showed good relationship between the tangent of the initial rate of the reaction and molar concentration of Cephalexin. The results of the proposed method are shown in the Tables 1-3.

Voltammetry

Cephalexin S-oxide in the reaction under study is formed through an electrophilic attack of the β -oxygen atom in the peroxide group of the peroxyacid to sulfur within 1 min, i.e., the time of observation. The scheme of Cephalexin oxidation by potassium caroate to give the respective S-oxide is presented (Fig 2, a).

Three peaks were recorded in the cathodic branch of the voltammograms of solutions: at -0.485 V (average sharp), at -0.800 V (average smooth), and -1.120 V (small smooth, Fig. 3). Considerable changes in peak height at -0.800 V was observed with changes in cephalexin concentration. It was chosen for analytical purposes.

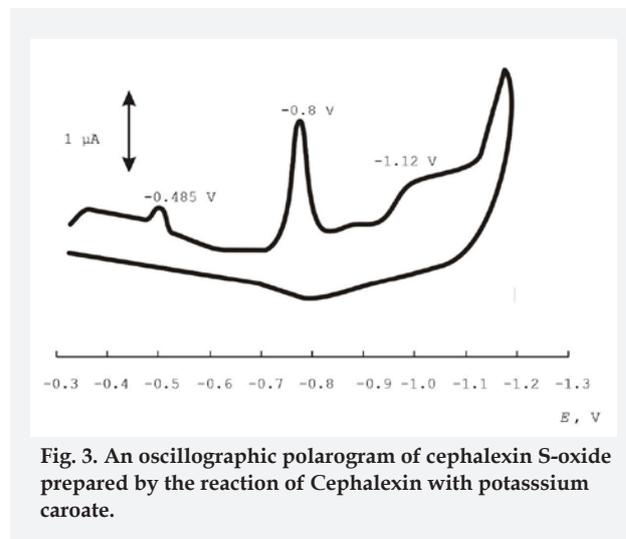


Fig. 3. An oscillographic polarogram of cephalexin S-oxide prepared by the reaction of Cephalexin with potassium caroate.

The calibration plot was obtained. The relation of current I , μA , at -0.800 V to the concentration of cephalexin c , mol L^{-1} , was approximated by the equation $I = (1.55 \pm 0.1) 10^4 c + (0.04 \pm 0.01)$, $r = 0.996$. The plot is linear in the range $(1-10) 10^{-5}\text{ M}$. This made possible the further determination by a reference method.

The data of Cephalexin determination in the pure substance is given in the Tables 1-3.

Iodometry

The proposed method is based on the S-oxidation reaction of Cephalexin by potassium caroate in acidic medium. The oxidation-reduction interaction was determined to be quantitative and stoichiometric: 1 mol of KHSO_5 per 1 mol of Cephalexin. The reaction product is cephalexin S-oxide (Fig.2, a).

The range of analytical procedure is the interval between 80% and 120% as recommended in SPhU.

Linearity is significant on the whole range of analyzed concentrations $(0.05-0.35\text{ mg mL}^{-1})$. The linear equation generated from the calibration curve was $Y = (1.025 \pm 0.13)X$ with a correlation coefficient $r=0.9997$. From this equation the content of cephalexin in the sample was calculated.

The detection limit (LOD) and the limit of quantification (LOQ) are less than 32% and do not influence on the quantitative determination.

The Limit of Quantitation (LOQ) is 0.05 mg mL^{-1} .

The advantages of the proposed method are ability of analytical determination of Cephalexin by the biologically active part of the molecule, mainly alicyclic Sulphur, good precision and accuracy. The method validated is faster compared to standard iodometric procedure (approx. 40 min).

The results of Cephalexin iodometric determination are given in the Tables 1-3.

DETERMINATION OF CEPHALEXIN IN SUSPENSION

According to the instruction the suspension was prepared as followed: 74 mL of water was added into the bottle with granules and stirred carefully. The solution was filtrated, 5.00 mL of filtrate was transferred into a 100 mL volumetric flask and diluted to the mark with twice-distilled water. Further analysis was performed as in the procedures described for iodometric, kinetic-spectrophotometric and voltammetric methods. The results obtained are summarized in Table 4.

CONCLUSIONS

Potassium caroate is used as analytical reagent for Cephalexin.

The proposed procedures were developed and validated. The obtained results show good agreement with the standard HPLC ($\delta < \text{RSD}$). The recovery percent ranged from 98.7 to 101.4. RSD from 1.42 to 3.0. The methods proposed are linear in a wide range: voltammetry $2-45\text{ }\mu\text{g mL}^{-1}$, kinetic-spectrophotometric method $1-50\text{ }\mu\text{g mL}^{-1}$, and iodometry $0.05-0.35\text{ mg mL}^{-1}$.

The iodometric method has the highest accuracy and precision $\text{RSD}=1.42\%$, $\delta=-0.19\%$.

The kinetic-spectrophotometric method is the most sensitive from the proposed ($\text{LOQ}=1.0\text{ }\mu\text{g mL}^{-1}$), it has higher accuracy and precision if compared to the voltammetric method ($F=0.45 < 6.39$).

All the developed procedures for the determination of Cephalexin in pure substance do not require elaborate treatment and expensive materials. The proposed methods are sensitive enough to enable determination of lower amounts of drug, these advantages encourage the application of proposed procedures in routine quality control of Cephalexin in industrial laboratories. Finally, these procedures provide advantages of improving selectivity, rapid, easy in performance and of a low cost.

REFERENCES

1. The 2010 Vidal Drug Compendium, 86th ed., French Health Products Safety Agency; 2010.
2. The State Pharmacopeia of Ukraine, suppl. 3, RIREG, Kharkiv; 2009.
3. British Pharmacopoeia, London: The Stationary office; 2009.
4. Patel SA, Patel NM, Patel MM. Spectrophotometric methods for the estimation of Cephalexine in tablet dosage forms. Indian J Pharm Sci. 2006; 68: 278-280.
5. Ni YN, Xiao WQ. Simultaneous kinetic spectrophotometric determination of cephalexin and trimethoprim in pharmaceutical preparation and human urine with the aid of chemometrics. Chinese Chem Lett. 2008; 19: 981-984.
6. Omara MA, Abdelmageeda OH, Attia TZ. Kinetic spectrofluorimetric determination of certain cephalosporins in human plasma. Talanta. 2009; 77: 1394-1404.
7. Elbashir AA, Ahmed SM, Aboul-Enein HY. New spectrofluorimetric method for determination of cephalosporins in pharmaceutical formulations. J Fluoresc. 2012; 22: 857-864.
8. Yao H, Tang Y, Li Y, Sun Y. Flow Injection Chemiluminescence Determination of Cephalosporin Antibiotics by Their

- Enhancing Effects on Luminol-Potassium Periodate System. *Anal Lett.* 2003; 36: 2975-2983.
- Patel SA, Patel NJ. Spectrophotometric determination of cephalixin using ninhydrin reagent in tablet dosage form. *IRJP.* 2011; 2: 123-126.
 - Chailapakul O, Aksharananandana P, Frelink T, Elinaga Y, Fujishima A. The electrooxidation of sulfur-containing compounds at boron-doped diamond electrode. *Sens Actuators B: Chem.* 2001; 80: 193-201.
 - Zuman P, Kapetanovic V, Aleksic M. Recent developments in electroanalytical chemistry of cephalosporins and cefamycins. *Anal Lett.* 2000; 33: 2821-2857.
 - Kharitonov SV. Ion-selective electrodes in medical drug determination. *Russ Chem Rev.* 2007; 76: 361-395
 - Karageorgou EG, Samanidou VF, Papadoyannis IN. Ultrasound-assisted matrix solid phase dispersive extraction for the simultaneous analysis of β -lactams (four penicillins and eight cephalosporins) in milk by high performance liquid chromatography with photodiode array detection. *J Sep Sci.* 2012; 35: 2599-607.
 - Baranowska I, Markowski P, Baranowski J. Simultaneous determination of 11 drugs belonging to four different groups in human urine samples by reversed-phase high-performance liquid chromatography method. *Anal Chim Acta.* 2006; 570: 46-58.
 - Malakhova NA, Ivanova AV, Kozitsina AN, Matern AI. Voltammetry in pharmaceutical analysis. *Drug Develop & Registr.* 2014; 2(7): 90-99.
 - Gallo ML, Campins FP, Sevillano CA. Comparison of several methods used for the determination of cephalosporins. Analysis of cephalixin in pharmaceutical samples. *J Pharm Biomed Anal.* 2002; 29: 405-423.
 - Validation of Analytical Procedures, Proceedinds of the International Conference on Harmonisation (ICH). Commission of the European Communities; 1996.

Table 1. Experimental data obtained in the recovery test for Cephalixin pure substance by voltammetric (VM), kinetic-spectrophotometric (KSM) and iodometric (IM) methods

Method	Added, g	Found, g	Recovery, %
VM	0.3654	0.3428	98.70
	0.2741	0.2719	99.20
	0.1827	0.1816	99.40
KSM	0.3654	0.3661	100.2
	0.2741	0.2766	100.92
	0.1827	0.1840	100.71
IM	0.3654	0,3705	101.41
	0.2741	0.2766	100.91
	0.1827	0,1823	99.79

Table 2. Analysis of Cephalixin pure substance accuracy determination

Method	Mean, %	RSD, %	N	$\bar{\delta}$, %	F	F=<6.39
VM	99.1	3.01	5	-1.78	1.07	
KSM	100.61	2.09	5	-0.29	0.45	
IM	100.71	1.42	5	-0.19	*	

*Reference method for F-data

Table 3. Characteristics of linear dependence for Cephalexin pure substance determination

Method	Concentration range, $\mu\text{g mL}^{-1}$	$Y = a \pm S_a + b \pm S_b \cdot X$	R	LOD $\mu\text{g mL}^{-1}$	LOQ $\mu\text{g mL}^{-1}$
VM	2-45	$I = (1.55 \pm 0.1)10^4 c + (0.04 \pm 0.01)$	0.996	1.0	3.0
KSM	1-50	$\text{tg}\alpha = (0,3621 \pm 0,013)10^{-2} c$	0.999	0.3	1.0
IM	50-350	$Y = (1.025 \pm 0.13)X$	0.999	16	50

Table 4. Analysis of Cephalexin drug quantitative determination

Method	Recovery $\pm D$, %	RSD, %	N	δ , %
VM	98.8 \pm 1.33	3.12	5	-0.46
KSM	100.9 \pm 1.07	2.17	5	+0.92
IM	99.37 \pm 1.93	1.56	5	-0.62