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Spectrophotometric Determination of Lisinopril Drug by Oxidative Coupling Reaction with 4-aminonaphthalene-1- sulfonic acid Reagent

Abdul Majeed Khorsheed Ahmed¹, Nashwan Omar Tapabashi², Noor Mansoor Ghaib Allah¹

¹ Chemistry Department, College of Education for Pure Sciences, Kirkuk University, Kirkuk , Iraq

² College of Pharmacy, Kirkuk University, Kirkuk, Iraq

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Corresponding Author:

Name: Abdul Majeed Khorsheed

E-mail:

noormansoor40@gmail.com

dr.majeed960@uokirkuk.edu.iq

Tel:

Introduction

Lisinopril is a class of angiotensin-converting enzyme (ACE) inhibitor drug used to treat high blood pressure, heart failure, and after heart attacks. It is commonly a first-line treatment for high blood pressure and also used in people with diabetes mellitus to avoid kidney problems. Lisinopril is taken by mouth. It can take up to four weeks for the full effect to occur [1]. In 2017, lisinopril was the most popular prescribed drug in the United States, having over 104 million prescriptions [2,3] In July 2016, a lisinopril oral formulation was licensed for use in the United States [4]. Lisinopril was invented in 1978 and allowed for medical use in the US [5]. The treatment of high blood pressure, cardiac obstruction, acute myocardial infarction (heart attack), and Diabetic Nephropathy is usually done with lisinopril [6]. Lisinopril leaves the body entirely unchanged in the urine, Lisinopril has a half-life of twelve hours [7]. Common side effects include headache, dizziness, exhausted feelings, cough, nausea, and rash. Low blood pressure, liver issues, higher blood potassium [8].

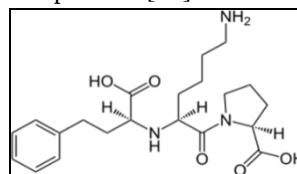
IUPAC Name is (2S)-1-[(2S)-6-amino-2-[[[(1S)-1-carboxy-3-phenylpropyl]amino]hexanoyl]pyrrolidine-2-carboxylic acid .

ABSTRACT

A simple, rapid and sensitive spectrophotometric method for determination of Lisinopril (LS) in both pure form and pharmaceutical preparations has been reported. The adapted technique based on utilization 4-aminonaphthalene-1-sulfonic acid as a Chromogenic reagent through an oxidative coupling reaction with Lisinopril and Potassium Peroxydisulfate (oxidation agent) in acidic medium to form green dye soluble product absorption maxima at 612 nm. Linearity was in the range 2–43 $\mu\text{g ml}^{-1}$. The values of molar absorption coefficient (ϵ), correlation coefficient and Sandel's index were found to be $10.3808 \times 10^3 \text{ L. mole}^{-1} \text{ cm}^{-1}$, 0.9995 and $0.00343 \mu\text{g.cm}^{-2}$ respectively. The average of recovery % was 100.067. Detection Limit and quantitative limit were $0.0343 \mu\text{g/ml}$ and $0.1143 \mu\text{g/ml}$. The proposed method has been successfully applied in the determination of preparations containing lisinopril.

Trade names are Zestril, Prinivil and Tensopril [9]. The pure powder of lisinopril is white to off-white. Lisinopril is soluble in water at room temperature, approximately 13 mg/L, [10] less soluble in methanol, and practically insoluble in ethanol. Melting Point is 148°C .

It releases poisonous vapors of nitric oxides when heated for decomposition [11].



Various analytical methods for determining lisinopril have been reported, including spectrofluorimetry [12], chemiluminescence [13], potentiometric titration [14], capillary zone electrophoresis [15], reverse phase-high-performance liquid chromatography [16] micellar electrokinetic chromatography [17] spectrophotometry [18] differential pulse voltammetry [19], and fluorescence [20]. In this work oxidative coupling method is proposed to determine Lisinopril in Pharmaceutical Preparations by coupling reaction with 4-aminonaphthalene-1-sulfonic acid in presence of

potassium peroxydisulfate as oxidation agent to form a green color product dissolved in water. Diverse techniques for assessing this substance and its properties have been used. Aim of the study is Determination of lisinopril in a spectrophotometric method through the oxidative coupling reaction

Experimental

Instruments:

Absorbance measurements were performed using T92 + Spectrophotometer.

Reagents:

All chemicals used in this study were of analytical grade.

Standard solution of Lisinopril (1000 μg / ml) was Prepared by dissolving 0.1013 g of lisinopril powder in a hot distilled water and the volume was completed to the mark in a 100ml volumetric flask with the same solvent. Solutions of lowest concentration of Lisinopril were Prepared by appropriate dilution.

Hydrochloric acid solution (1 M) was prepared by diluting 8.5 ml of concentrated hydrochloric acid of (11.8 N) with distilled water and the volume was completed to 100 ml in a volumetric flask.

The oxidizing agent Potassium peroxydisulfate (2×10^{-2} M): this solution was prepared by dissolving 0.27 g of potassium peroxydisulfate powder in a volume of distilled water and diluted to the mark in a 100 ml volumetric flask with the same solvent.

4-aminonaphthalene sulfonic acid (1×10^{-2} M) solution: This solution was prepared by dissolving 0.223g of 4-aminonaphthalene sulfonic acid in a volume of hot distilled water and diluted to in volumetric flask to 100 ml with the same solvent.

Pharmaceutical preparations of Lisinopril

Each tablet contains 10 mg. The solution of the tablets was prepared by crushing 10 tablets (4.052) gm, 0.1g is taken and dissolved in hot distilled water in a volumetric flask of 100 ml and the solution is filtered and the sediment is washed several times with distilled water, then the volume complete to the mark with distilled water to obtain a solution at a concentration of 1000 μg / ml. 25 ml of the prepared solution was diluted by distilled water to the mark in a 100volumetric flask to obtain concentration of 250 μg / ml.

Preliminary study:

When adding 1.5 ml of reagent 4- Aminonaphthalene sulfonic acid to 1 ml of the oxidizing agent potassium peroxydisulfate and when adding 1.5 ml of a solution of Lisinopril drug in the presence of 1 ml of HCl 1M, a green product was obtained which absorbs at 612 nm against the blank solution

Results and Discussion

Study of the optimum reaction conditions:

Experiments were performed using the method by which the lisinopril drug was coupled with 4-aminonaphthalene sulfonic acid in presence of Potassium peroxydisulfate as oxidizing agent in an acidic medium, and a green-colored solution gave the

highest absorption wavelength of 612 nm versus blank solution.

Effect of the best coupling Reagent:

1.5 ml of each of the other reagents solutions 1×10^{-2} M and 1 ml of the oxidizing agent solution of 1×10^{-2} M potassium peroxydisulfate were mixed with 1.5 ml of a solution of lisinopril of 250 μg / ml; 1 ml of 1 M hydrochloric acid and the results are shown in Table (1).

Table 1: Effect of coupling reagent type

Reagent 2×10^{-2} M	Variable	Absorbance	λ_{max}
4-amino naphthalene-1-sulfonic acid	SB	0.635	612
	BW	0.073	500
O-aminophenol	SB	0.213	495
	BW	0.082	430
P-aminophenol	SB	0.276	489
	BW	0.097	415
Resocinol	SB	0.196	515
	BW	0.088	420
4- aminobenzen sulfonic	SB	0.317	485
	BW	0.085	439

From the above table it is clear that the reagent 4-aminonaphthalene -1- Sulfonic acid gave the highest absorption of the colored product at 612 nm against the blank solution. The blank solution did not show any absorption at this wavelength.

Effect of the coupling Reagent quantity:

The effect of the coupling reagent quantity was studied by taking different volumes (0.4- 2.6) ml of 0.01 M of 4- aminonaphthalene sulfonic acid in the presence of 1 ml of 0.01M potassium peroxydisulfate, 1.5ml of 250 μg /ml lisinopril and 1 ml of 1M hydrochloric acid solution .It was observed that the volume of 1.5 ml of the coupling reagent solution gives the highest absorption value, so this volume was chosen in the subsequent experiments. The results are shown In the table (2)

Table 2: Effect of the coupling reagent quantity

ml of Reagent $M1 \times 10^{-2}$	Absorbance	
	BW	SB
0.4	0.072	0.362
0.6	0.080	0.397
0.8	0.092	0.485
1.0	0.188	0.556
1.2	0.208	0.599
1.5	0.244	0.635
1.8	0.181	0.605
2.0	0.097	0.562
2.2	0.030	0.476
2.4	0.032	0.303
2.6	0.03	0.296

Oxidizing agent optimization:

Several oxidizing agents were used at a concentration of 1×10^{-2} M, each with a volume of 1 ml, and 1.5 ml of the reagent solution 4-aminonaphthalene sulfonic acid was added 1.5 ml of 250 μg / ml lisinopril and 1 ml of 1M hydrochloric acid solution ,in a volumetric Flask of 25 ml after which the absorption was measured for each sample against the blank solution and it was noted that the best oxidizing agent is potassium peroxydisulfate , which gives the highest absorption of the colored product at the wavelength of 612 nm and the results are shown in Table (3).

Table 3: Test from best oxidizing agent

Oxidizing agent 1×10 ⁻² M	Absorbance		λ max(nm)
	Blank	Sample	
Potassium Iodate	0.091	0.274	475
Potassium peroxydisulfate	0.040	0.633	612
Ammonium per Sulphate	0.073	0.198	500
Ammonium ferric Sulphate	0.039	0.104	410

Selection of the acid used for coupling: 1 ml, 1M of different acids (strong and weak) was used and its effect on the absorption of the formed product was studied and the results are shown in the table (4).

Table 4: Effect of the Acid used in the coupling

Acid Solution Used 1 M	HCl	H ₂ SO ₄	HNO ₃	CH ₃ COOH
Absorbance	0.634	0.602	0.453	0.189

The concentration of a used lisinopril was 250 µg/ml, 1.5 ml

Effect of the amount of acid used:

Various quantities of the acid (HCl) were added it was found that 1 ml gives the best absorption (pH value was 2.3), so the volume of 1 ml was adopted in the subsequent experiments and the results are shown in Table (5).

Table 5: Effect of quantity of the acid used

ml of 1 M HCl	Absorbance		Ph
	BW	SB	
0.2	0.108	0.491	3.00
0.4	0.109	0.445	2.73
0.6	0.098	0.541	2.58
0.8	0.103	0.614	2.46
1.0	0.104	0.635	2.37
1.2	0.102	0.601	2.26
1.4	0.095	0.524	2.24
1.6	0.082	0.385	2.15
1.8	0.071	0.318	2.11
2.0	0.103	0.276	2.08

Table 7: Effect of Temperature

Temperature °C	15	20	25	30	40	50	60	70	80
Absorbance	0.633	0.634	0.635	0.635	0.632	0.628	0.622	0.515	0.285

Stability of the reaction product:

When studying the stability of the product by measuring the absorption of the formed product against the blank solution at different time intervals at a temperature of 25°C, to know the stability of the

Sequence of Addition:

The sequence of addition of the solutions used sometimes has an effect on the absorption of the colored product. Therefore, a number of experiments were conducted with a different sequence of additions, noting that all the volumes and concentrations of the substances used were the same in all cases. It was noted from the results obtained in table (6) that the order (III) gives the highest absorption of the colored product, so it has been adopted in subsequent experiments:

Lisinopril (D), reagent solution 4-aminonphthalein sulfonic acid (R), Potassium peroxydisulfate solution (O), and hydrochloric acid (A)

Table 6: Effect of order sequence of addition

Order Number	Order of addition	Absorbance	
		BW	SB
I	D+ O+ A+R	0.041	0.538
II	A +D +R+ O	0.020	0.423
III	O + R + D+A	0.052	0.635
IV	R +A +D +O	0.037	0.392

Effect of Temperature:

The temperature of mixture solution was varied at the range (15-80) Celsius. Maximum and stable absorption of the formed colored product solution was observed at the temperature range (20-30) Celsius thus 25 °C was chosen to be the optimal temperature for the reaction mixture. The results are shown in the table (7).

formed product, we take three different volumes of the lisinopril solution (250µg/ml), so the final concentrations were (10 20, 30µg/ml) and results are shown in Table 8.

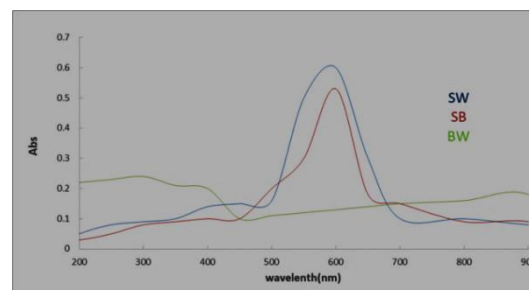
Table 8: Effect of time on absorption of the formed product

µg/ml of ls	Absorbance / min . Standing time								
	5	10	15	20	25	30	40	50	60
10	0.628	0.628	0.629	0.630	0.630	0.631	0.630	0.632	0.631
20	0.633	0.631	0.632	0.633	0.632	0.632	0.633	0.634	0.635
30	0.634	0.634	0.635	0.634	0.633	0.634	0.635	0.632	0.634

The results in Table No. (8) show that the formed product is stable for an hour, and this time is Practically enough to complete many measurements.

Ultimate absorption spectrum:

The final absorption spectrum was measured after optimum conditions were established. 1 ml of 10⁻² M potassium peroxydisulfate, 1.5ml of 10⁻² M 4-aminonaphthalene sulfonic acid and 1.5 ml of 250 µg / ml lisinopril solution, with 1 mL of 1M HCl were mixed in 25 ml volumetric flask and diluted to the mark with distilled water. The absorption of the green product was measured against the blank solution and found to absorb at 612 nm, the spectrum shown in Figure (1).

**Fig. 1: Ultimate absorption spectrum**

SW: the absorption spectrum of lisinopril solution versus distilled water, SB: the absorption spectrum of lisinopril solution versus blank solution, BW: the absorption spectrum of the blank solution versus distilled water

Approved working method and calibration curve:

After fixing the optimum conditions for the method, the standard curve was prepared as follows:

Increased volumes (0.5 - 7.5) ml of lisinopril solution of 250 µg/ml were added to a series of 25ml volumetric flasks containing 1.5 ml of the reagent 4-aminonaphthalene sulfonic acid 1×10^{-2} M and 1 ml of potassium persulfate and 1 ml of 1 M HCl solution, then complete the volume to the mark (25ml) with distilled water and then measure the absorbance of all solutions against the blank solution at the wavelength of 612 nm. Figure (2) represents the standard curve that follows Beer's law in the limits of concentrations (2 - 43) µg / ml of lisinopril solution.

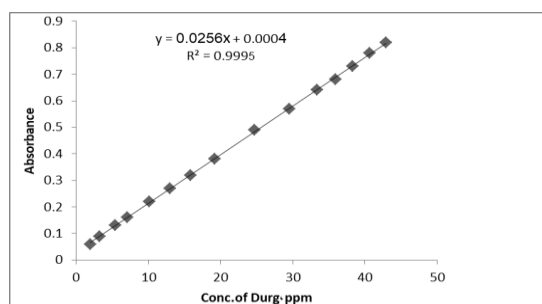


Fig. 2: Calibration curve

Table 9: The accuracy and precision of the method

Conc. Of LS(taken) (µg/ml)	Conc. of LS (measured) (µg/ml)	RE%	Recovery %	Average of Recovery %	RSD %
10	10.1	1	101.0	100.067	0.3954
18	17.91	-0.5	99.5		0.5367
33.5	33.4	-0.298	99.70		0.9063

*Each value averages five readings

Detection Limit

The detection limit of this method was calculated at the wavelength of 612 nm, by measuring the absorption of the lowest concentration (2 µg/ ml) taken from the calibration curve value averages seven readings under the same conditions.

Table 10: Detection Limit

Conc. Of ls µg/ml	$\bar{X}$	S	D.L. µg/ml	Q.L. µg/ml
2	0.098	0.000748	0.04579	0.1526

The nature of the formed product

To know the nature of the formed product and the ratio of the drug's binding to the reagent, continuous changes method (**Job's method**) and the molar ratio method were applied. In both methods, the concentration of each of the lisinopril solution and the reagent solution is equal to which is 1×10^{-2} molar. In the Job method, a series of 25 ml volumetric flask was placed and different volumes of the drug solution ranging from (1-8) ml. were placed into the flasks containing the rest of the additives were at the optimum sizes according to the method of work, and then diluted to the mark with distilled water and then measure the absorption of these solutions at 612 nm against blank solution. and Figure (3) shows that the ratio is 1: 1

The value of the molar absorption coefficient of the method was 10.3808×10^3 L. mole⁻¹ cm⁻¹ and the Sandel significance was 0.0390625 µg.cm⁻², and the value of the correlation coefficient is 0.9996. This indicates that the standard curve is of good linear and accuracy.

Accuracy and precision:

The accuracy of the method represented by Relative Error, RE% and Recovery Percentage (Rec%) were calculated. The precision of the method represented by Relative Standard Deviation RSD% was calculated for three concentrations (10, 18, 33.5) µg/ml. The results obtained in Table (9) shows that the method has good accuracy and precision.

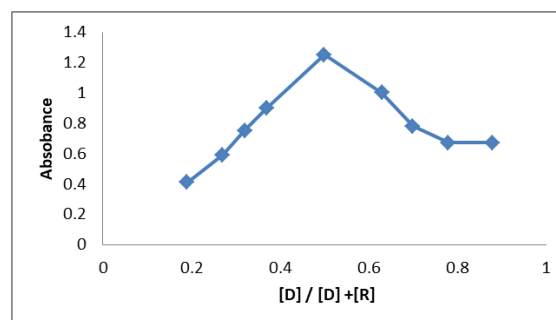


Fig. 3: Job's method for determination of Lisinopril

To ensure that the binding ratio between Lisinopril and the 4-aminonaphthalene-1-sulfonic acid reagent is 1: 1, I used the molar ratio method. 2 ml, 250 µg/ml of the drug solution was placed in a series of 25 ml volumetric flasks containing different volumes of the reagent solution (0.2 -2) ml, 1×10^{-2} M, with the remaining additives at the optimum sizes, and diluted with distilled water to the point of the mark, and then measure the absorption of these solutions at the wavelength of 612 nm against the blank solution and it was found that the molar ratio is consistent with the method of continuous changes and Figure (4) shows that the ratio It is 1: 1 between the drug lisinopril and the reagent 4- amino-naphthalene sulfonic acid

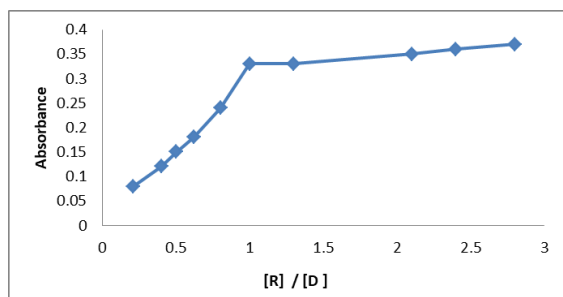
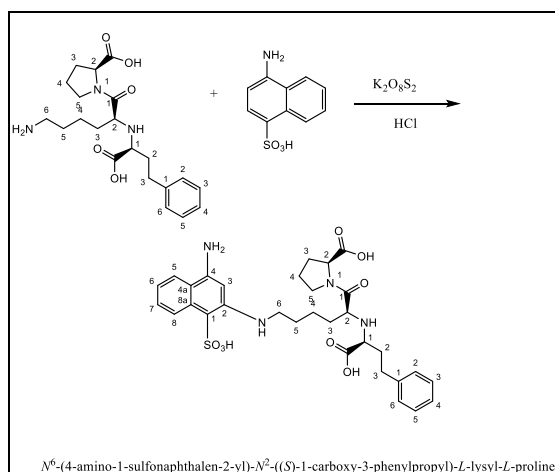


Fig. 4: Molar ratio method

Accordingly, the proposed equation will be as follows. The reagent first oxidizes and then reacts with Lisinopril in an acidic medium



Applications:

The method can be applied to the following pharmaceutical preparations containing lisinopril
_ Lisinopril STADA tablet

Direct method:

Three different concentrations were taken from a solution of a preparation (Tablets) indicated in the preparation in paragraph 10, 24, 36 µg/ ml and the solutions were treated with the same steps when preparing the calibration curve and then measured the absorption of them at the wavelength of 612 nm against the blank solution and the average of five readings was calculated For each as well as the recovery account and RSD The results are shown in Table No. (11)

Table 11: The direct method

Conc. Of lisinopril	Conc. of measured	RE%	Recovery %	Average of Recovery %	RSD %
10	10.019	0.19	100.19	100.025	0.4079
24	23.618	-1.59	98.40		0.0761
36	36.4	1.05	101.1		1.0369

The results of the above table show that the proposed method has succeeded in appreciation the pharmaceutical preparations that contain them. The value of the average of recovery was 100.025, for tablets.

Standard Additions Method:

The standard addition method was used to demonstrate the efficiency and accuracy of the proposed method in estimating lisinopril in pharmaceutical preparations. The method included adding constant quantities (1.0, 2.3 ml), 250 µg / ml of pharmaceutical preparations solution prepared , in two series of volumetric flasks of 25 ml. increased volumes (1, 2, 3, 4, 5 ml) of 250 µg / ml, standard solution were added and treated as in the method of Calibration graph .

Table 12: Standard additions method for determination of Ls in tablets

Amount taken µg/ml	Amount Measured	Recovery %
7	6.8	98.60
9.3	9	98.83

From the results shown in the table (12), it is obvious that the standard addition method is in agreement with the direct method.

Conclusions

An easy, simple and highly sensitive spectroscopic method has been developed for determination of lisinopril based on the oxidative coupling reaction of the drug with the 4- Amino naphthalene sulfonic acid reagent in the presence of the oxidizing agent potassium peroxydisulfate to form a green color product , stable and soluble in water, gives the highest absorption at the wavelength of 612 nm and obeys Beer's law in the range 2-43 µg/ml of Ls. The molar absorption coefficient was 10.3808×10^3 L/ mol.cm, Sandel index $0.0390625 \mu\text{g} / \text{cm}^2$ and correlation coefficient (0.9995). The method was successfully applied in estimating lisinopril in pharmaceutical preparations (tablets) with a recovery of (100.067)%.

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التقدير الطيفي لدواء اللايسينوبريل بتفاعل الاقتران التأكسدي بوجود الكاشف 4-أمينو نفتالين حامض السلفونيك

عبدالمجيد خورشيد احمد¹, نشوان عمر رشيد², نور منصور غيب الله¹

¹قسم الكيمياء, كلية التربية للعلوم الصرفة, جامعة كركوك, كركوك, العراق

²كلية الصيدلة, جامعة كركوك, كركوك, العراق

الملخص

يتضمن تطوير طريقة طيفية سريعة وحساسة لتقدير كميات ضئيلة من ليزينوبريل في محلول مائي حامضي، تعتمد هذه الطريقة على الاقتران التأكسدي للعقار مع الكاشف 4-امينو نفتالين سلفونيك اسيد بوجود العامل المؤكسد بوتاسيوم بير سلفات لتكوين ناتج اخضر اللون ذائب بالماء مستقر، يعطي اعلى امتصاص عند الطول الموجي 612 نانومتر ويطيع قانون بير بحدود (2- 43 مايكروغرام/مل)، وبلغت الامتصاصية المولارية 10.3808×10^3 لتر/مول. سم، ودلالة ساندل 0.0390625 مايكروغرام/ سم² وحد الكشف 0.0343 مايكروغرام/مل، والحد الكمي 0.1143 مايكروغرام / مل، الانحراف القياسي النسبي بين (0.3954 - 0.9063 %) ومعدل الاسترجاعية 100.067 %, معامل التقدير 0.9995 وطبقت هذه الطريقة المقترحة بنجاح لتقدير ليزينوبريل في مستحضراته الدوائية.