Spectrophotometric Determination of Enalapril maleate by Using1, 2 Naphthaquinolinc-4-Sulphonate Sodium Reagent

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Abstract

In this study, simple and sensitive spectrophotometric method for the determination of Enalapril maleate (ENP) in pharmaceutical formulations was reported. The proposed method was based on the reaction between (ENP) and 1, 2-naphthoquine-4-sulphonate (NQS) at alkaline medium (pH= 12) to form deep red product. Beer's law was obeyed in the range of (5-47.5) µg.mL^{-1} of Enalapril maleate at maximum wavelength of 518 nm. Under optimized reaction conditions, linear regression equation of the calibration curve was y = 0.0179 x + 0.1888 (µg.mL^{-1}) with a linear correlation coefficient of 0.9988. The limit of detection (LOD) and limit of quantification (LOQ) were found to be 0.3351 µg.mL^{-1} and 1.1173 µg.mL^{-1}, respectively. The method was successfully applied to the determination of (ENP) in pharmaceutical formulations.

Keywords: Spectrophotometric; Enalapril maleate (ENP), pharmaceutical formulation; 1-2- naphthoquine-4-sulfonate (NQS)

DOI Number: 10.14704/nq.2022.20.10.NQ55975 **NeuroQuantology 2022; 20(10): 10014-10030**

Introduction

Enalapril maleate is chemically described as (2S)-1-[(2S)-2-[[(1S)-1- (Ethoxycarbonyl)- 3-phenylpropyl] amino] propanoyl] pyrrolidine-2-carboxylic acid (Z)-butenedioate fig (1). Enalapril maleate is a prodrug which metabolized rapidly in the liver to ethyl ester of a long- acting enalaprilat which inhibits angiotensin-converting enzyme (ACE) in human subjects and animals. ACE is a peptidyl dipeptidase that catalyzes the conversion of angiotensin I to the vasoconstrictor substance, angiotensin II. Angiotensin II also stimulates aldosterone secretion by the adrenal cortex. The beneficial effects in hypertension and heart failure appear to result primarily from suppression of the renin-angiotensin-aldosterone system. Inhibition of ACE results in decreased plasma angiotensin II, which leads to decreased vasopressor activity and to decrease aldosterone secretion [1]. A literature survey was conducted and several methods were reported for the determination of enalapril maleate including the following methods: Uv-Visible spectrophotometry [2-3], HPLC [4-5], HPTLC [6] and LC-MS/MS [7].

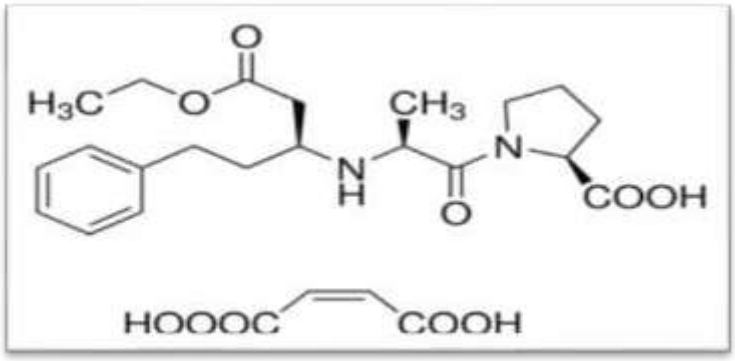

Figure (1) The structure of Enalapril maleate

Medical uses

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Enalapril is used to treat hypertension, symptomatic heart failure, and asymptomatic left ventricular dysfunction.[8] ACE-inhibitors (including enalapril) have demonstrated ability to reduce the progression and worsening of existing chronic kidney disease in the presence of proteinuria/microalbuminuria (protein in the urine, a biomarker for chronic kidney disease) [9]. This renal protective effect is not seen in the absence of proteinuria/microalbuminuria, including in diabetic populations[10]. The benefit has been particularly demonstrated in patients with hypertension and/or diabetes, and is likely to be seen in other populations (although further studies and subgroup analyses of existing studies are needed) [11].It is widely used in chronic kidney failure.[12]. Furthermore, enalapril is an emerging treatment for psychogenic polydipsia. A double- blind, placebo-controlled trial showed that when used for this purpose, enalapril led to decreased water consumption (determined by urine output and osmolality) in 60% of patients.[13].

Side effects

The most common side effects of enalapril include increased serum creatinine (20%), dizziness (2–8%), low blood pressure (1–7%), syncope (2%), and dry cough (1–2%). The most serious common adverse event is angioedema (swelling) (0.68%) which often affects the face and lips, endangering the patient's airway. Angioedema can occur at any point during treatment with enalapril, but is most common after the first few doses [8] .

Mechanism of action

Normally, angiotensin I is converted to angiotensin II by an angiotensin-converting enzyme (ACE). Angiotensin II constricts blood vessels, increasing blood pressure. Enalaprilat, the active metabolite of enalapril, inhibits ACE. Inhibition of ACE decreases levels of angiotensin II, leadingto less vasoconstriction and decreased blood pressure [8]. 1,2-Naphthoquinone-4-sulfonic acid sodium salt)(NQS): Synonyms: beta-Naphthoquinone-4-sulfonate sodium salt; Sodium 3,4- dihydro-3,4-dioxo-1-naphthalenesulfonate; Sodium beta-naphthoquinone-4-sulfonate; Sodium 1,2- naphthoquinone-4-sulfonate Fig. (2).

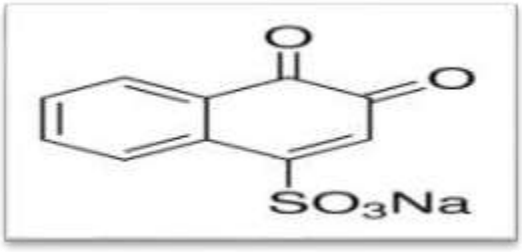

Figure (2). Chemical structure (1,2-Naphthoquinone-4-sulfonic acid sodium salt) (Folin's reagent)(NQS)

1, 2-naphthoquinone-4-sulfonic sulfonate (NQS) has been used as a chromogenic reagent for the spectrophotometric determination of many pharmaceutical amines.[14,15] .It is a popular spectrophotometric reagent due to its efficient reactivity with both primary and secondary amines, and high reaction rate [16]. NQS proved to be a useful and sensitive analytical derivatizing agent for spectrophotometric analysis of pharmaceuticals bearing a primary or secondary amino group [17]**.** Many researchers have intensively used NQS as a chromogenic reagent to determine amines and the results obtained showed that the reaction products have the general structure shown in fig. (3) [18,19]**.**

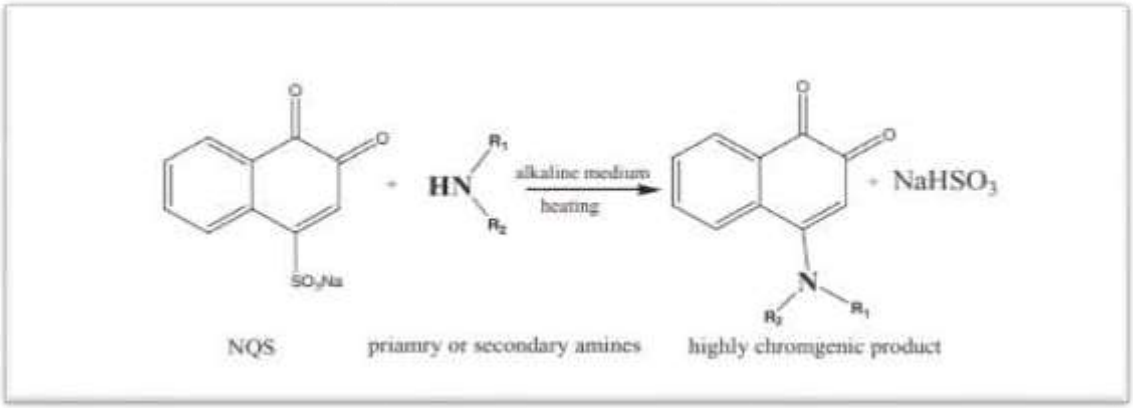

Figure (3): Reaction scheme of amines with NQS

Experimental

Apparatus

Absorbance was carried out by using 6100 PC UV- visible spectrophotometer, Shimadzu, Japan (Double beam), with quartz cells of 1 cm optical path length, pH meter was used for pH measurements, analytical balance and water bath.

Material and reagent

Enalapril maleate we r e ob t a i ne d from Development Company, (agent of Hyber chem), used as received, the purity of which was 98%. The preparation of the solution of ENP involved dissolving 0.055 g Enalapril in the alkaline solution(0.1M) NaOH and then complete by Deionized water in 100 mL volumetric flask. A solution of 0.5% (w/v) NQS was prepared by dissolving 0.5 g in distilled water, transferred into a 100 mL volumetric flask and diluted to the mark with Deionized water and mixed well. The solution was freshly prepared and protected from light. during use; buffer solution of pH 12.

Preparation of standard and sample solution

The standard stock solution of (250 µg.mL^{-1}) was prepared from (0.025) g Enalapril dissolved in 50 Deionized water , then transferred into 100 mL volumetric flask, diluted to the mark with the same solvent and mixed well. This stock solution was further diluted to obtain working solutions in the range of (5-47.5) µg.mL^{-1}.

Solution of Pharmaceutical Preparations of Enalapril maleate

Ten tablets were weighed and finely powdered from each type of tablets separately. An accurately weighed portion of the powder equivalent to (0.025) g of Enalapril maleate which depends on the type of tablets that be used, it was dissolved in 10 mL of distilled water and added 1 ml from (0.1 M) NaOH. After that it was filtered to separate then dissolved components, and transfer to a 100mL volumetric flask and to diluted to the mark with distilled water. Later, the suitable amount of each record solution was taken and treated in the same conditionsthat were used in the based way of working to find a concentration depending on a calibration curve .

10017

Procedure

1 mL of (250) µg.mL^{-1} Enalapril was transferred into 10 mL volumetric flask; 1 mL of 0.5% (w/v) NQS was added and added 1 ml from buffer solution of pH =12 (NaOH / NaHPO4). The reaction was completed to volume by deionized water, and the resulting solution was measured at 518 nm against reagent blank treated similarly without added the drug. The Job's method of continuous variation and mole ratio [52] were employed to evaluate the stoichiometric ratio of formation reaction. Master equimolar (4×10^{-3} mol) aqueous solution of Enalapril and NQS were prepared. Series of 10 mL portions of the master solution of Enalapril and NQS were made comprising different complementary proportions (0.1:0. 9, …0.9:0.1, inclusive) of reagent and ENP under optimal condition at wavelength of 518 nm and the ratio of Drug to reagent at 1:1.

Results and DiscussionAbsorption spectra

The absorption spectrum of Enalapril (ENP) was recorded against water. It was found that ENP exhibits a maximum absorption peak (λ_{max}) at 271 nm. The reaction between ENP and NQS was performed, and the absorption

spectrum of the product was recorded against the reagent blank (Figure 6).It was found that the product was colored exhibiting (λ_{max}) at 518 nm, and the (λ_{max}) of NQS was 342 nm.The (λ_{max}) of ENP- NQS derivative was red-shifted, eliminating any potential interference. Therefore, the measurements were carried outat 518 nm.

The UV-visible spectrum of ENP , Figure (5), showed a band at wavelength (271) due to the electron transition(n→ π*). As for the spectrum resulting from the interaction of the drug with the NQS reagent, Figure (6), it is located at the wavelength (518 nm), which is a beam that goes back to the electronic transition (n→ π*). We note from the results that the beam (n→ π*) has a red shift in the resulting spectrum.

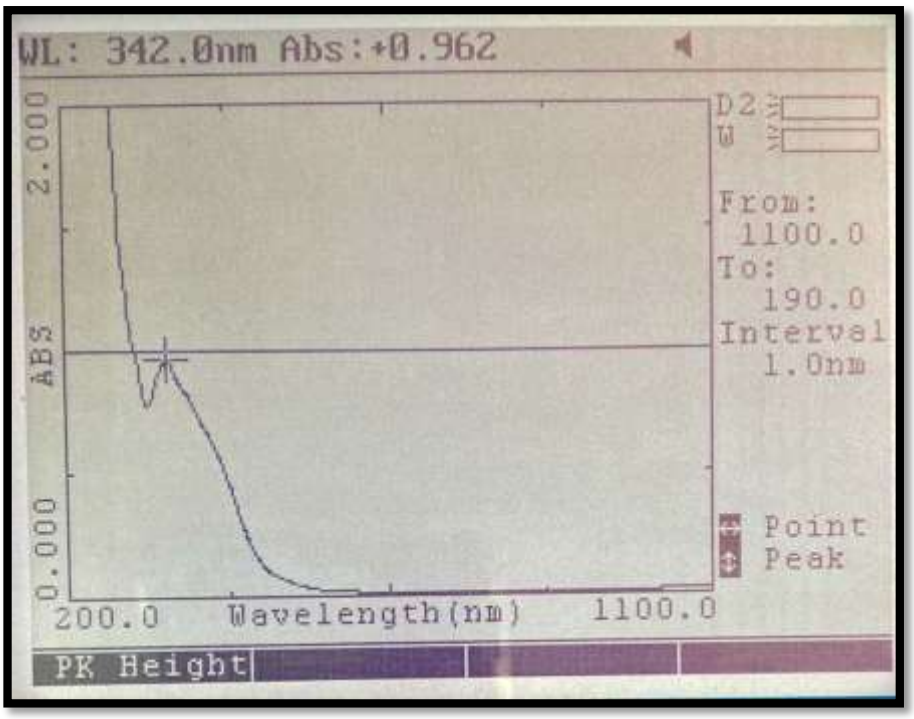

Figure (4) : UV-Visible Spectrophotometer for reagent 1, 2 Naphthoquinone-4-sulfonic acid sodiumsalt (NQS)

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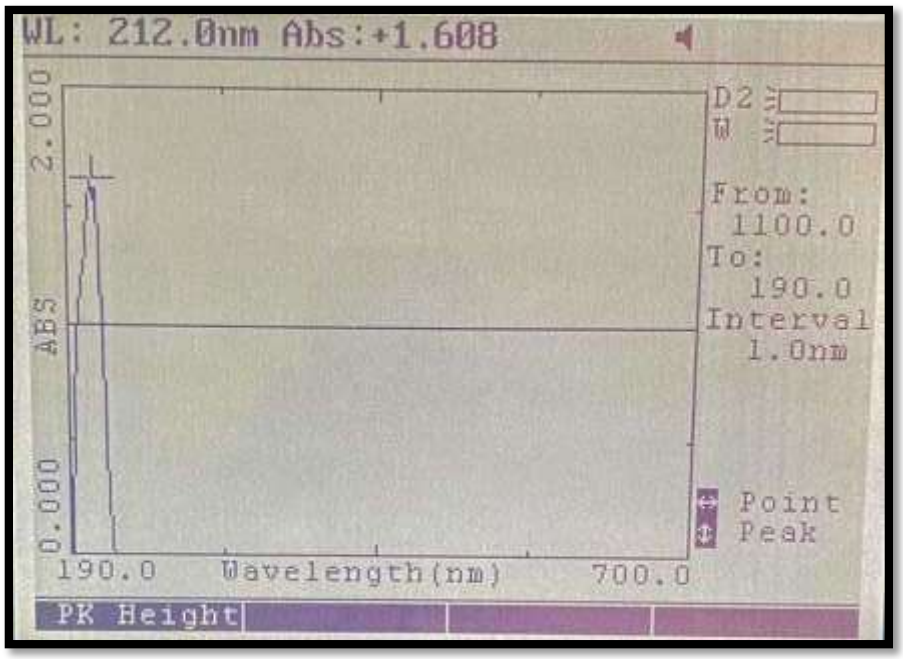


Figure (5) : The UV-visible spectrum of pure ENP

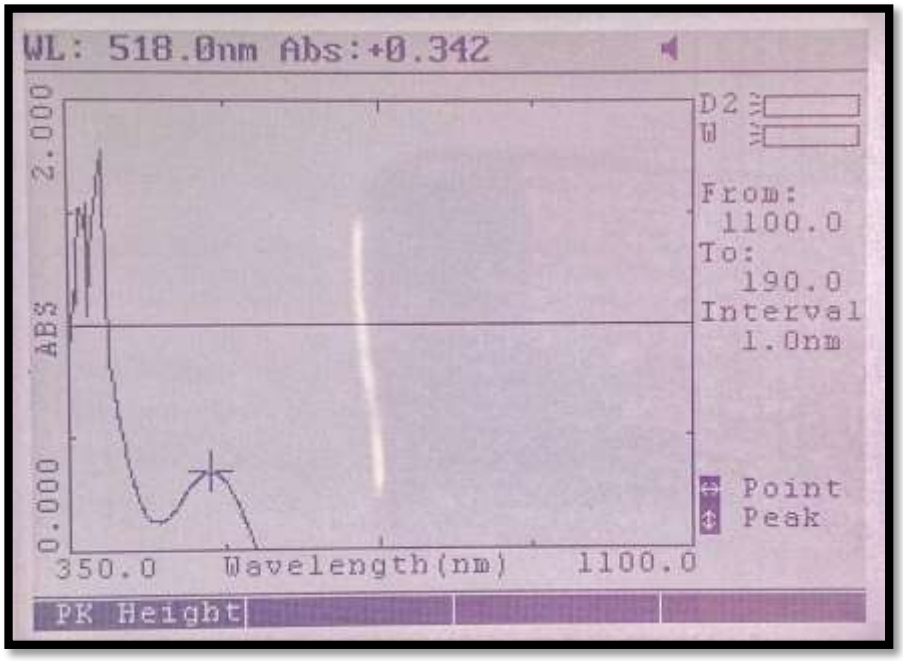


Figure (6): UV-visible spectrum of the reaction product of ENP with the NQS reagentOptimization of the reaction conditions

Effect of volume of the Drug

The study investigated the effect of the volume of ENP by taking different amounts of ENP and measuring the absorption intensity of solution using the wavelength of ENP. The highest absorption intensity was detected at the best volume of Drug ENP at 1 mL (Figuer. 7).

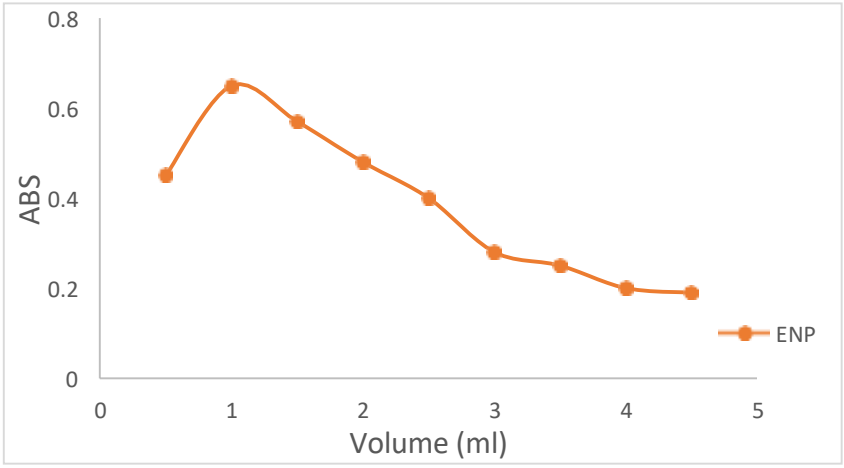


Figure (7): Effect of Volume Drugs

Effect of (NQS) Concentration

The studying of (NQS) concentrations revealed that the reaction was dependent on (NQS) reagent. The highestabsorption intensity was attained at (NQS) concentration of 0.5 (w/v) %, (0.019M) the higher concentration of (NQS) (1.0 w/v%) had no effect on the absorption values in the Drug (ENP) as shown in Figure (8).

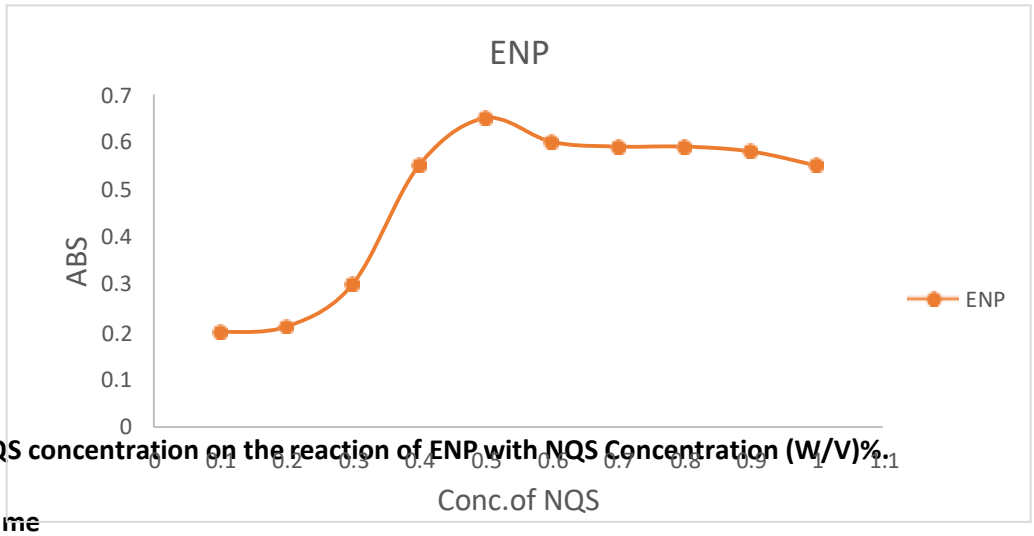


Figure (8): Effect of NQS concentration on the reaction of ENP with NQS Concentration (W/V)%.

Effect of Reagent Volume

The effect of the volume of reagent on the intensity of absorption of the resulting dye was studied by taking different volumes of reagent with a concentration of 0.5% (w/v) NQS for ENP. The highest absorption intensity was detected at 1 mL (Figure 9).

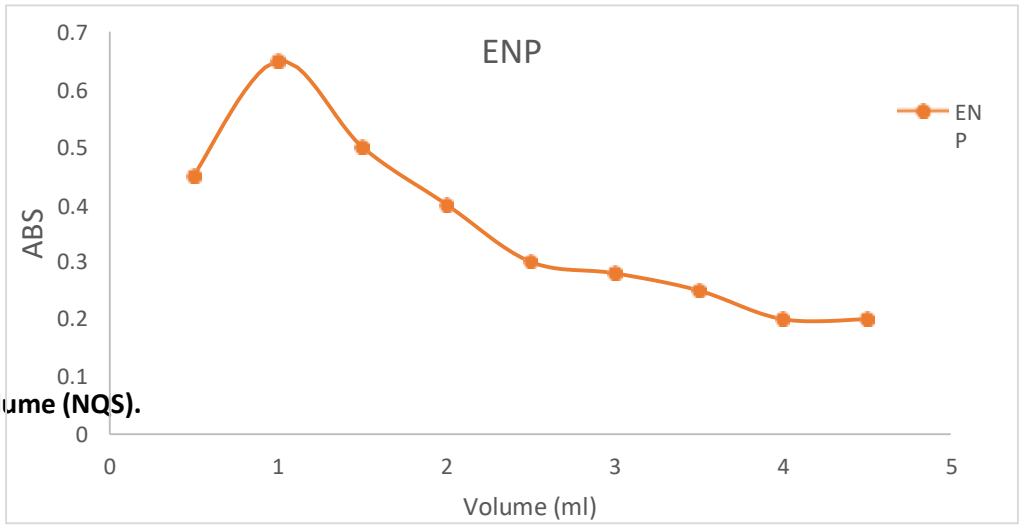


Figure (9): Effect of Volume (NQS).

Effect of pH of the buffer solution

After the installation of condition of the best in the past experiences were study effect of pH on the reaction Drugs ENP with (NQS) was examined by a varying pH form (6.0 - 13.0). As shown in Figure (10), the absorbance of the product is low at pH 9.0, indicating that the Drug has a difficulty to react with (NQS) in pH 9.0. This was possibly due to the existence of the amino group of Drugs in the form of salt, thus it loses its nucleophilic substitution capability. The maximum readings were attained at, the value of the of drug ENP was equal 12.

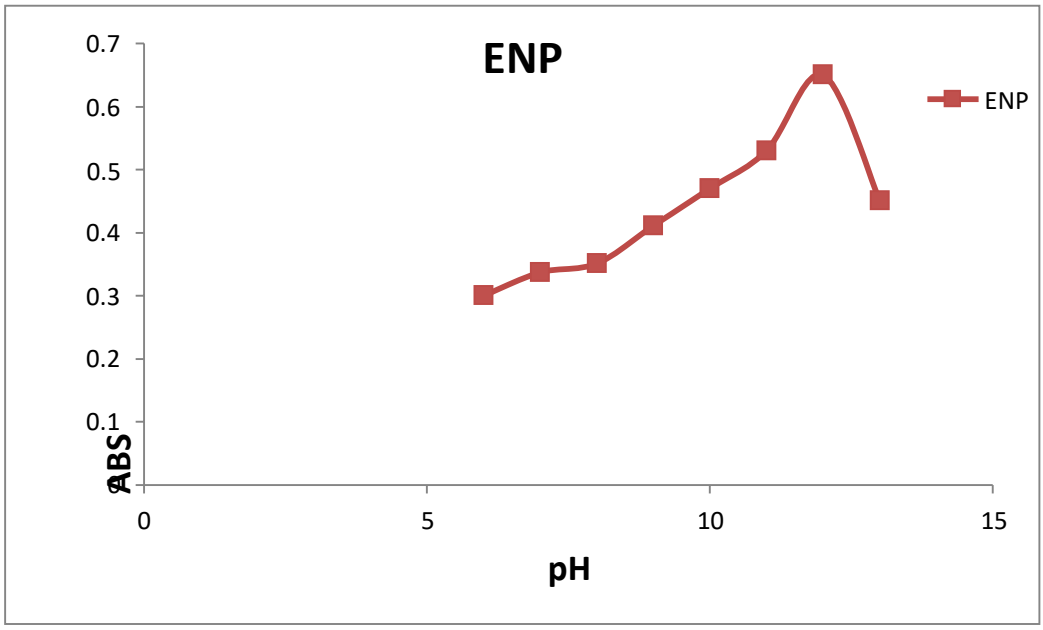


Figure (10) : Effect of pH on the reaction of ENP and with NQS.Effect of amount of the buffer

The effect of the amount of buffer solutions of drugs (ENP) on the absorbance of the reaction product was also studied. figure (11) shows that the absorbance of the reaction product enhances rapidly with the rise of the amount of buffer solutions until to (1mL) to ensure the highest absorbance.

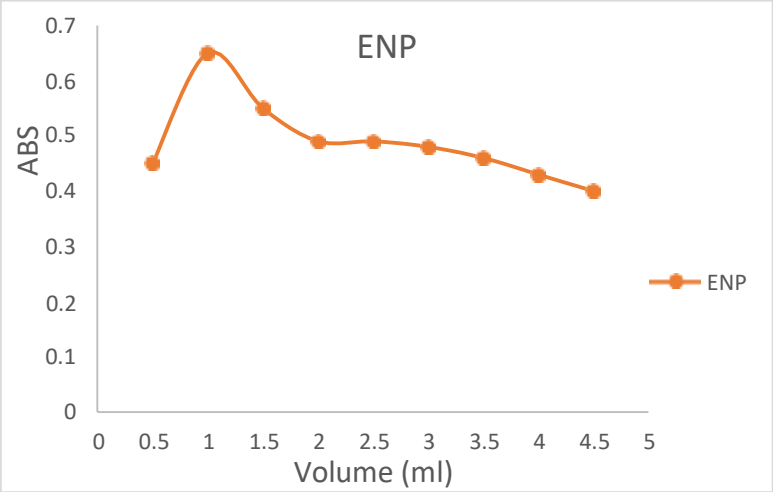


Figure (11): Effect of Volume (pH)

Effect of Reaction Time

For the purpose of studying the effect of time in the stability of the mixture consisting of the (NQS) and Drug, the absorbance of the reaction product was determined at a different time as in figure (12). The absorbance of the reaction product was measured after standing for different time under the other optimal condition . The best reaction times of the mixture are detected depending on the high absorbance from drugs (ENP) are 15 min.

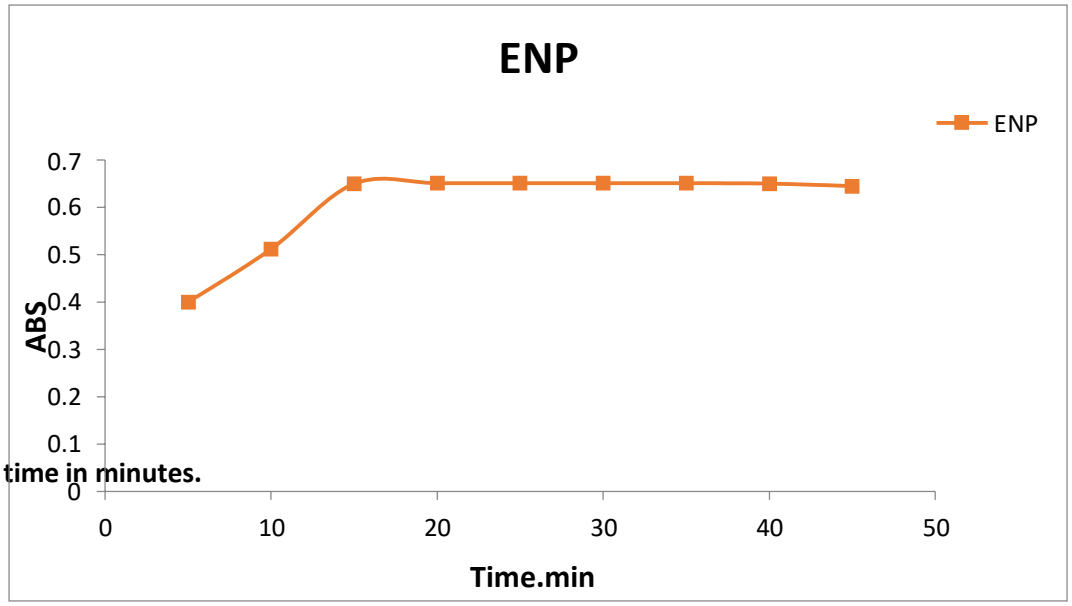


Figure (12): Effect of time in minutes.

Effect of temperature

The effect of temperature on the stability of NQS and Drug reaction was detected. Fig. (13) reveals the besttemperature for reaction was 25 °C when ENP gave the highest absorption.

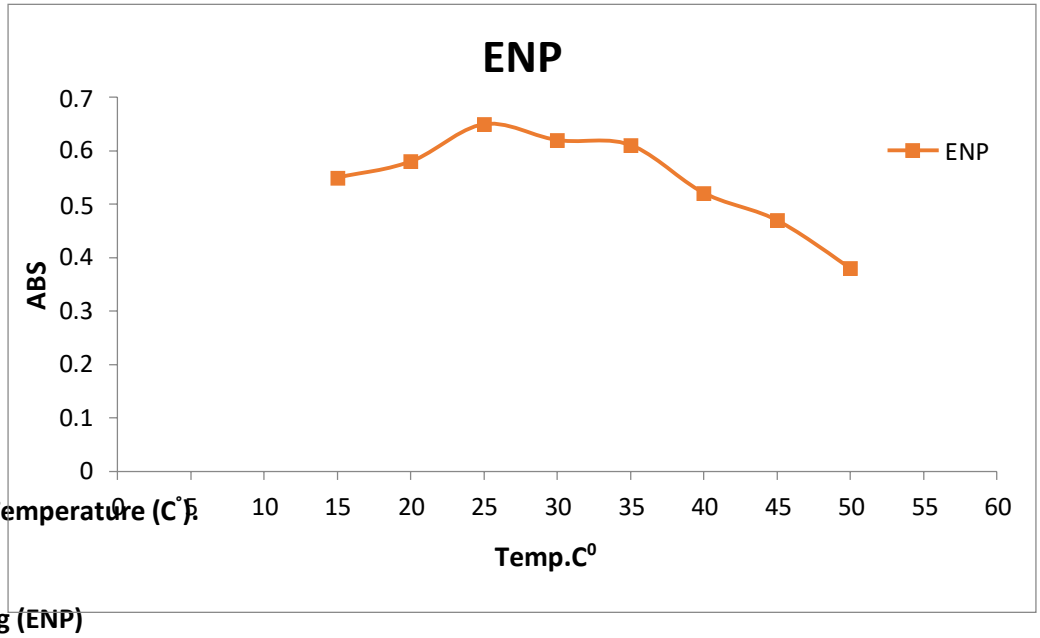


Figure (13): Effect of Temperature (C°).

Calibration Curve Drug (ENP)

Figure (14) shows the standard calibration curve for the estimation of the drug (ENP). It turnsout that it obeys the Beer's Law between the range of (5-47.5) µg.mL^{-1} under the best conditions at the maximum wavelength of (518) nm and the correlation coefficient (R^2=0.9988). As for the value of was (Ɛ) molar absorptivity, it was equal to (0.8815×10^4) L.mol^{-1}.cm^{-1}. The sandell's sensitivity was calculated by calculating the coefficient of specific absorption (a) of the following relationship and was equal to (0.5596) µg.cm^{-2}**.** The value of molar high absorptive and the sandell's sensitivity make this analytical method preferred for drug (ENP) determination at low concentrations.

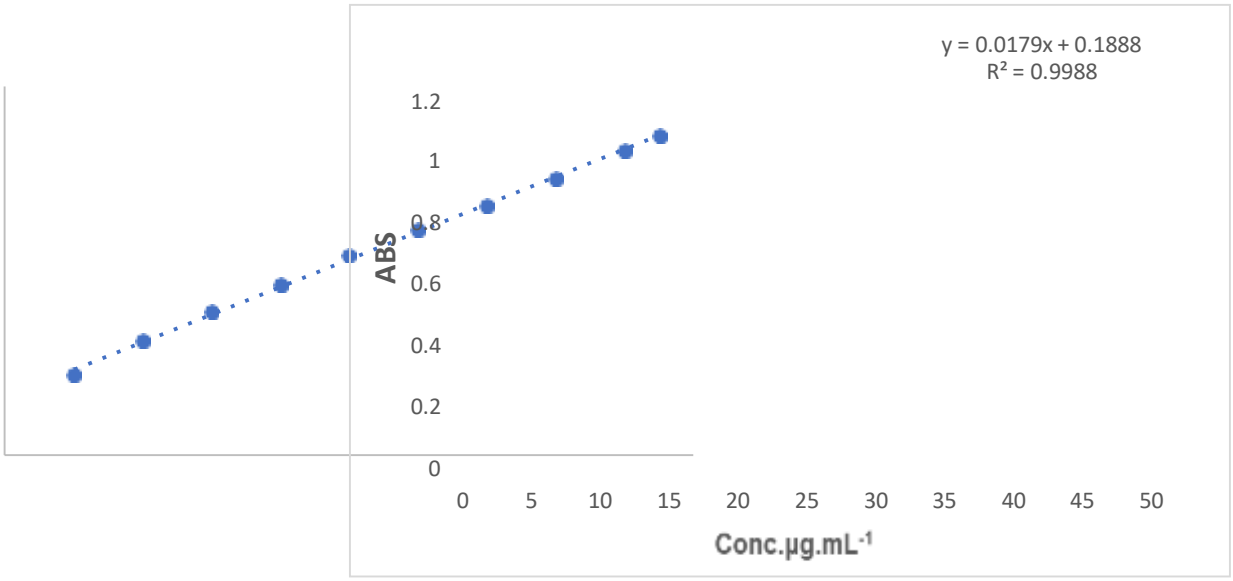


Figure (14): Calibration Curve Drug (ENP).

Table (1): Analytic parameter for (ENP) determination.

parameter Value	ENP
Beer's law limit (µg.mL^{-1})	(5-47.5)
Molar absorptivity (L.mol^{-1}.cm^{-1})	0.8815×10^{4}
Sandell's sensitivity(µg.cm^{-2})	0.5296
Detection limit (µg.mL^{-1})	0.3351
Quantitation limit (µg.mL^{-1})	1.1173
Determination coefficient(R^2)	0.9988
Slope (b)	0.0179
Intercept (a)	0.1888

Estimation the Composition of the ProductDrug (ENP)

In the present work, the stoichiometric ratio of Drug (ENP) and (NQS) was investigated applying the continuous variation (Job's methods) and the mole ratio methods to evaluate the formation of the reaction between the reagent and Drug (ENP) under the optimal condition at a wavelength (518) nm ,as seen in figure (15). It was found that the drug forms a product with (NQS) in the ratio (1:1).

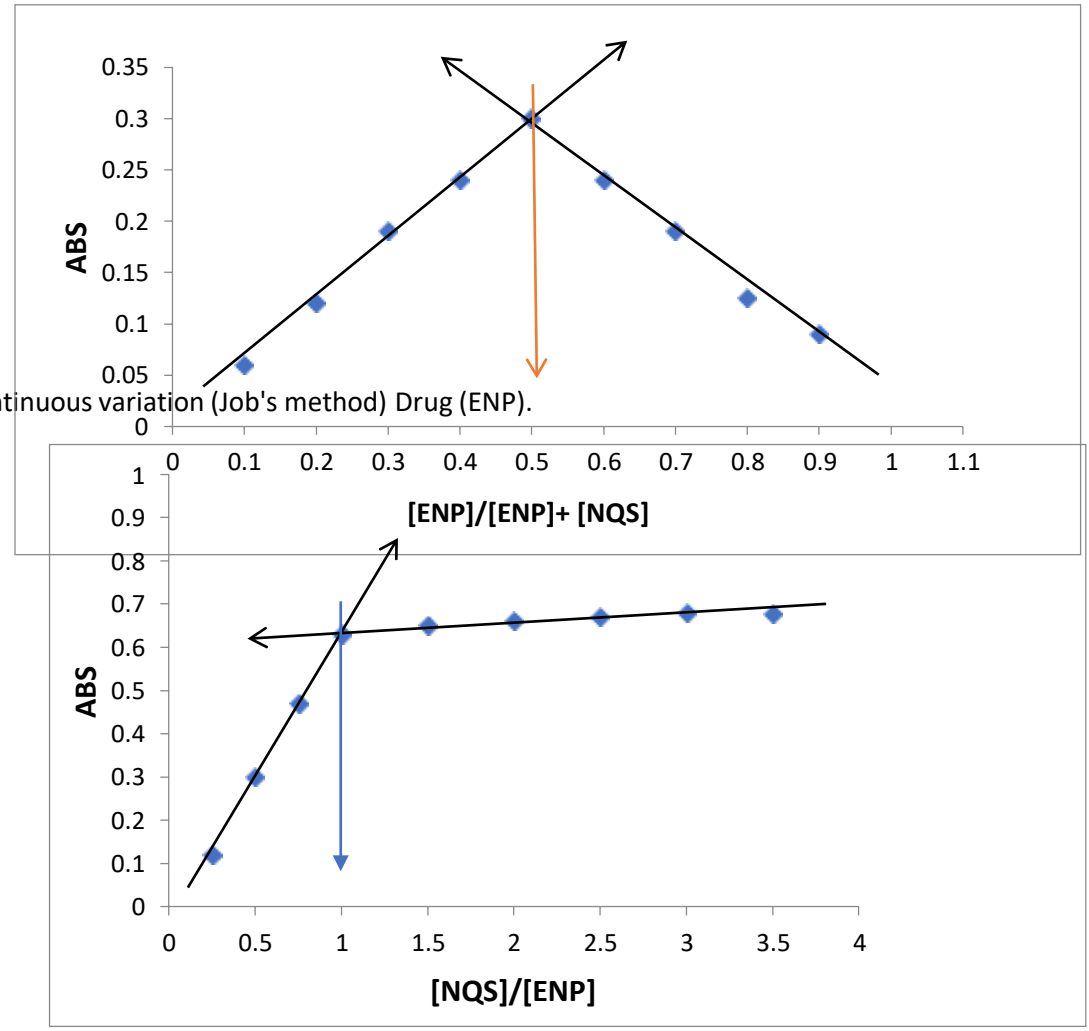
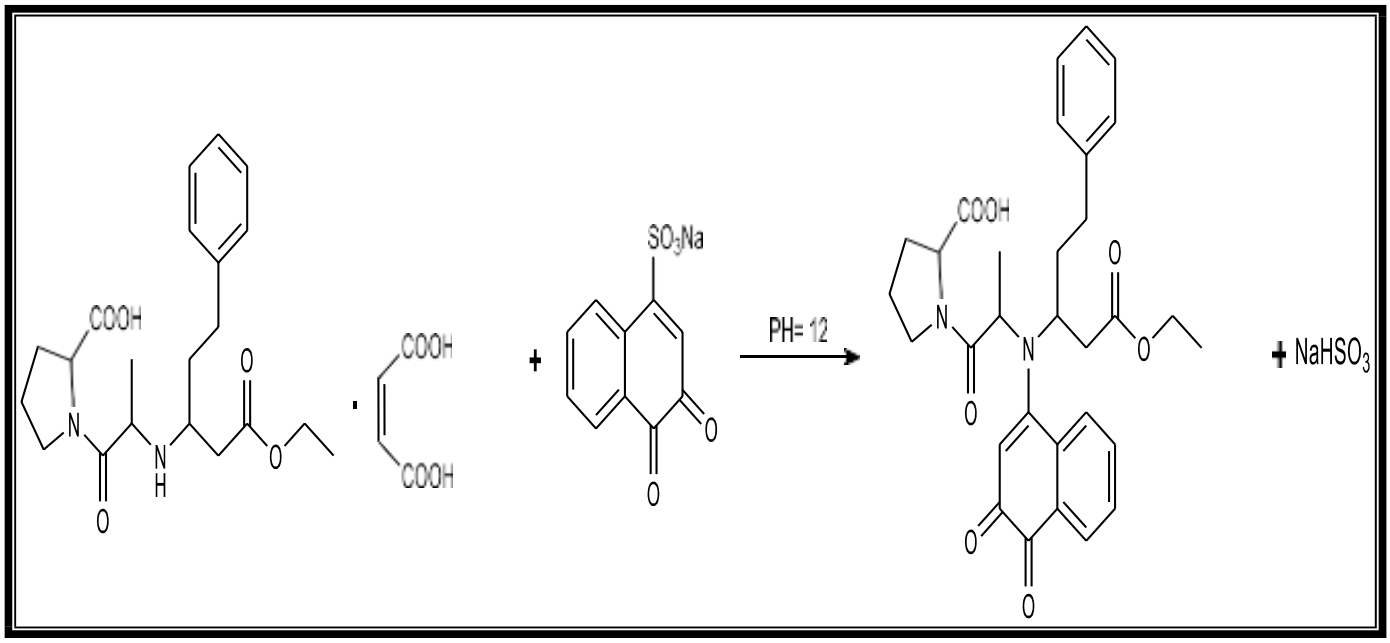


Figure (15) the continuous variation (Job's method) Drug (ENP).

Figure (16) mole ratio Drug (ENP)



Scheme 1. Reaction Suggest Enalapril maleate (ENP)

Table (2): Relative errors and recovery as parameters expressing accuracy of methods todetermine Drugs.

Values	(ENP)
wavelength(nm)	518
Conc.	25µg.mL^{-1}
$\bar{x}$	8.640
R.S.D %	0. 380%
Error %	-1.46%
%Recovery	98.54

Application of the proposed method to analyse ENP drug formulations

The proposed method indicated the analytic purpose of determining ENP in tablets . The results are listed in Table (3) to determine Drug in pharmacological formula by spectrophotometric methods.

Table (3): Analytical applications (ENP).

Preparations containing (ENP)	Concentration \ (µg.mL^{-1})			
	Present	**Found**	**Error %**	**Recovery %**
Enapril maleate Enapril 10 mg Niksan	15	14.94	-0.33	99.67
	25	24.97	-0.12	99.88
	45	45.02	+0.04	100.04

Conclusions

The analytical method was easy, simple and inexpensive for the determination of antihypertensive drugs (MPT, ENP, BIF and ATV) spectrophotometrically; also they have the high precision, accuracy and selectivity. The alkaline medium was good condition for the reaction of antihypertensive drugs (MPT, ENP, BIF and ATV) with (NQS) reagent. The reaction with (NQS) obeys Bear's-Lambert law in different reagent for each antihypertensive drugs. The method has no effective of interference which found in each pharmaceutical formulation for each. This method has stable product after period of time which are (15 min) for each antihypertensive drugs (ENP, BIF, ATV) and time (30 min) for (MPT) with (NQS). The stoichiometry for the method by (NQS) reagent for antihypertensive drugs equal (1:1) for (ENP, BIF) and (1:2) for (MPT, ATV). High value of stability constant for all products of method by (NQS) reagent while low degree of disintegration. Calculation the thermodynamic for the two methods for determination of the drugs (MPT, ENP, BIF and ATV). Study of kinetic parameter ΔG, ΔH and ΔS for determination of the drugs (MPT, ENP, BIF and ATV). The results of calculating the thermodynamic values showed that drug preparation reactions are endothermic reactions, and they are reactions automatic. Assay of metoprolol by measuring blood concentration in a single form afterwards Stabilization, linking the blood

measurement to the quality of the brand products you use. Assessment Lipid Profiles in Patients with CVD Group and Control Group by Using statistics to identify hyperlipidemia patients who use atorvastatin calcium with those who do not use it.

References

1- Logoyda, L., Abdel-Megied, A. M., Kondratova, Y., Trofimenko, O., Korobko, D., & Dakhym, I. (2018). Development and validation of HPLC method for the simultaneous determination of enalapril maleate in present of their impurities: Application to tablet analysis. *International Journalof Applied Pharmaceutics*, *10*(1), 98–102. https://doi.org/10.22159/ijap.2018v10i1.22805

2- Manoranjani, M., & Karuna, K. K. (2011). UV-visible spectroscopic estimation and validation of enalapril maleate in bulk and pharmaceutical dosage forms. *Journal of Research in Pharmaceutical and Biomedical Sciences*, *2*(4), 1651–1656.

3- Sowjanya, G., Gangadhar, P., Ramalingeswara Rao, P., Subrahmanyam, P., & Suresh, P. (2012). Simultaneous Uv. spectrphotometric estimation of enalapril maleate and hydrochlorothiazide in tablets. *Journal of Chemical and Pharmaceutical Research*, *4*(7), 3483–3488.

4- Roškar, R., Simončič, Z., Gartner, A., & Kmetec, V. (2009). Stability of new potential ACE inhibitor in the aqueous solutions of different pH. *Journal of Pharmaceutical and Biomedical Analysis*, *49*(2),295–303.

5- Lima, D. M., dos Santos, L. D., & Lima, E. M. (2008). Stability and in vitro release profile of enalapril maleate from different commercially available tablets: possible therapeutic implications. *Journal of Pharmaceutical and Biomedical Analysis*, *47*(4–5), 934–937.

6- Mennickent, S. C., Rivas, C., Vega, M. A., & De Diego, M. G. (2013). A stability-indicating HPLC method for quantification of enalapril maleate in tablets. *Journal of the Chilean Chemical Society*,*58*(2), 1737–1740.

7- Danafar, H., & Hamidi, M. (2015). Liquid chromatography–tandem mass spectrometry (LC-MS) method for the assignment of enalapril and enalaprilat in human plasma. *Pharmaceutical and Biomedical Research*, *1*(3), 47–58.

8- Ravishankar, K., & Kiranmayi, G. V. N. (2021). Enalpril in Covid 19: A Possible Role Beyond Hypertension". *J Pharmacy and Drug Innovations, 2 (5)*; DOI : http. *Doi. Org/03.2020/1.1022*.

9- Xie, X., Liu, Y., Perkovic, V., Li, X., Ninomiya, T., Hou, W., Zhao, N., Liu, L., Lv, J., and Zhang, H. (2016). Renin-angiotensin system inhibitors and kidney and cardiovascular outcomes in patients with CKD: a Bayesian network meta-analysis of randomized clinical trials. *American Journal of Kidney Diseases*, *67*(5), 728–741.

10- Bangalore, S., Fakheri, R., Toklu, B., & Messerli, F. H. (2016). Diabetes mellitus as a compelling indication for use of renin angiotensin system blockers: systematic review and meta-analysis of randomized trials. *Bmj*, *352*.

11- Mishima, E., Haruna, Y., & Arima, H. (2019). Renin-angiotensin system inhibitors in hypertensive adults with non-diabetic CKD with or without proteinuria: a systematic review and meta-analysis ofrandomized trials. *Hypertension Research*, *42*(4), 469–482.

12- He, Y., Feng, L., Huo, D., Yang, Z., & Liao, Y. (2013). Enalapril versus losartan for adults with chronic kidney disease: A systematic review and meta-analysis. *Nephrology*, *18*(9), 605–614.

13- Greendyke, R. M., Bernhardt, A. J., Tasbas, H. E., & Lewandowski, K. S. (1998). Polydipsia inchronic psychiatric patients: therapeutic trials of clonidine and enalapril. *Neuropsychopharmacology*, *18*(4), 272–281.

14- Li, Q.-M., & Yang, Z.-J. (2007). Spectrophotometric determination of aminomethylbenzoic acid using sodium 1, 2-naphthoquinone-4-sulfonate as the chemical derivative chromogenic reagent. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, *66*(3), 656–661.

15- Darwish, I. A. (2005). Kinetic spectrophotometric methods for determination of trimetazidine dihydrochloride. *Analytica Chimica Acta*, *551*(1–2), 222–231.

16- Abdullah, H. J., & Qassim, B. B. (2022). Development and Validation CFIA/MZ System as

*e*ISSN1303-5150

a Green Method for Determination of Thiol Drug (D-PEN). *Egyptian Journal of Chemistry*, *65*(1), 1–2.

17- Kucukkolbasi, S., Bilber, O., Ayyildiz, H. F., & Kara, H. (2013). Simultaneous and accurate determination of water-and fat-soluble vitamins in multivitamin tablets by using an RP-HPLC method. *Química Nova*, *36*(7), 1044–1051.

18- Hashimoto, Y., Endo, M., Tominaga, K., Inuzuka, S., & Moriyasu, M. (1978). Quantitative analysis of phenethylamine derivatives by thin layer chromatography. Determination of psychotropic drugs and ephedra bases. *Microchimica Acta*, *70*(5), 493–504.

19- Altigani, A. M. N., & Elbashir, A. A. (2014). Spectrophotometric method for determination of Primaquine in pharmaceutical formulations via derivatization with 1, 2-Naphthoquinone-4-sulfonate. *Austin Journal of Analytical and Pharmaceutical Chemistry*, *1*, 1019.

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