ISSN: 0975 -8542

Journal of Global Pharma Technology

Available Online at: www.jgpt.co.in

RESEARCH ARTICLE

Spectrophotometric Determination of Atenolol via Oxidative Coupling Reaction with 2, 4 - Dinitrophenyl Hydrazine

Raed M. Hussein*, **Intidhar D. Sulaiman**

Department of Chemistry, College of Education for Pure Science (Ibn Al-Haitham), University of Baghdad, Baghdad, Iraq.

***Corresponding Author: Raed M. Hussein**

Abstract

An correct and sensitive spectrophotometric methodology has been developed for the determination of beta-adrenergic blocker (ATN) in pure and dose forms. the strategy is predicated on the oxidization of 2,4-dinitrophenylhydrazine (2,4-DNPHz) by metallic element per iodate than coupling with beta-adrenergic blocker (ATN) in a calescent medium to create actable, Yellow colored , soluble dye with a most absorption at 403 nm. The variables that have an effect on the completion of reaction are fastidiously optimized. Beer's law is obeyed over the concentration vary of (2-30 µg.mL-1) with molar absorption factor of (3.515×104 L.mol-1.cm1).The limit of detection was (0.07575µg.mL-1) and Sandal's sensitivity price was 0.0350 µg.cm-1. The projected methodology has been applied with success to the determination of antihistamine in pharmaceutical preparation.

Introduction

ß- Blockers or (ß-adrenergic antagonists) are a gaggle of medicine wide employed in the treatment of vas diseases (CVD), namely, blood vessel cardiovascular disease, viscos arrhythmias, and cardiopathy in addition as different sorts of pathologies like anxiety or eye disease [1]. The therapeutic effects of beta blockers are ordinarily explained by their capability to dam the betaadrenoceptors, preventive access of the endogenous agonist's monoamine neurotransmitter and visions attractive.

Atenolol, 4-(2-hydroxy-3-isopropylaminopropoxy) painkiller (Fig. 1) that in medicine is understood as a ß-blocker and is wide employed in the management of cardiovascular disease, cardiopathy, viscos arrhythmias and infarction [2]. Determination of beta blocker. Some reported ways suffer from one or a lot like crucial dependence on acid/pH condition, heating and/or extraction step, use of use of organic solvents, longer contact time, less stable colored species and costly chemicals as indicated in table one. For these reasons, develop a brand new easy spectrophotometric technique for the determination of ATN in their pharmaceutical indefinite quantity forms victimization eco-friendly chemicals and free from the utilization of organic solvents. The drug is official within Indian accumulation [3] and within the British accumulation [4]. many analytical ways are reported for the termination of beta blocker in human plasma, urine, or pharmaceutical preparations, like superior liquid natural action [5, 9], gas natural action [10], liquid natural action [11, 12].

Different techniques embrace voltammetry [13, 15], natural action [16, 17], luminescence [18, 20], Spectrofluorimetric [21], ultraviolet and visual spectrophotometry [22, 27]. The aim of the current work is to recommend a straightforward and sensitive spectrophotometry procedure for the determination of Coinciding in pure indefinite quantity forms. The strategy is predicated on AN oxidization of two, 4-dintrophenyhydrazin by metal per iodate and reaction with carbamazepine in alkali medium to make a colored product.

Additionally, the reaction conditions were studied one-factor-a time to supply AN optimized analytical response.

Experimental

Instruments

APG instrument, UV-visible spectrophotometer model T80 (U.K) with 1cm matched quartz cells was used for the absorbance measurements. Sartorius BL 210 S electronic balance was used for weighing the samples.

Materials and Methods

All chemicals used were of analytical reagent grade and were obtained from BDH and Pancreas. Atenolol standard powder was kindly provided by the State Company for Drug Industries and Medical Appliances, Samara-Iraq (SDI).

Atenolol Stock Solution (1000 µg.mL^{-1})

The stock resolution of (ATN.) was ready by dissolving Associate in Nursing accurately weighed zero.1000 g of pure drug in 10ml of grain alcohol and therefore the volume was created up to the mark in200mL meter flask with grain alcohol. The stock resolution was protected against lightweight and hold on at.

Atenolol Working Solution (200µg.mL^{-1})

Prepared by diluting 20mL of the stock answer to 100mL during a volumetrically flask with ethanol.

2,4-dinitrophenyl hydrazine solution (2, 4-DNPHz) (1×10^{-3}M)

Prepared by dissolving 0.0198g of 2,4-DNPHz in 3mL of focused oil of vitriol and also the volume was created up to the mark in100mL volumetrically flask with water.

Potassium Periodate Solution (3×10^{-3}M)

Prepared by dissolving zero.069g of KIO4 in a very appropriate volume of water and therefore the volume were created up to the mark in 100mL meter flask.

Sodium Hydroxide Solution (~4M)

Prepared by dissolving 16.000g of NaOH in a suitable volume of distilled water and the volume were made up to the mark in 100mL volumetric flask.

Atenolol Tablets Solution (1000µg.ml-1)

The content of ten tablets was accurately weighed and grinded into fine powder then mixed well and a mean weight was calculated. associate quantity of the powder adore 0.4456g (containing 0.1g of the drug Atenolol) of beta blocker fifty mg, beta blocker a hundred mg, VASCOTEN100 mg .respectively was accurately and severally weighed, dissolved in termly plant product and stirred for 10 min to confirm complete dissolution of the drug, then transferred into fifty cubic centimeter meter flask and diluted to the mark with plant product to induce 1000 µg.mL-1 (ATN.).

The answer was filtered by victimization what man paper No.41 to avoid any suspended or covert material before use. Operating resolution (200µg.mL-1) was freshly ready and analyzed by the suggested procedure.

General suggested procedure for standardization in an exceedingly series of 10mL meter flasks, 1mL of two×10-3M 2, 4-DNPHz and 1mL of 3×10-3M metallic element per iodate were side to every flask. The ensuing change product was as well as (ATN.) by adding 1mL aliquots of the quality resolution containing (20 -300) µg followed by 1mL of 1M hydroxide to every flask with shaking. Once 10min, the solutions were creating up to the mark with water, mixed well and left to square for 10min. The absorbance of buff colored chromogenic was measured at 403 nm against their agent blank.

Results and Discussion

Absorption Spectra for Primary Test

The primary take a look at for this methodology concerned oxidization of 2,4-dintrophenyhydrazin with metal amount Greek deity and reaction with (ATN.) in alkalescent medium to create a colored product. The take a look at was done by adding 1ml of 200µg.ml-1 (ATN.), 1mL of 1×10-3M two,4-DNPHz, 1mL of 3×10-3M metal amount Greek deity, and so 1mL of 1M hydrated oxide in 10mL volumetrically flask with shaking. The contents were diluted to the mark with H2O. The absorbance and λ max of the colored product was measured against the chemical agent blank. Figure (1) shows that the most absorption was obtained at a wavelength of 403nm.

Optimization of Reaction Variables

The various parameters associated with the colored product formation are studied by variable the parameters one at a time and dominant all others mounted and optimum conditions are elect.

Effect of 2, 4-DNPHz Concentration

Of the concentration of two, 4-DNPHz on the absorbance of the colored product was investigated within the vary between (6×104-4×10-3)M Figure (2). It absolutely was found that the utmost absorbance of the raw sienna color was achieved with 2×10-3M of the chemical agent. Higher than this worth a decrease in absorbance was ascertained. Therefore, 1mL of 2×10-3M was used throughout the following work.

Effect of Potassium Periodate Concentration

The studies of potassium periodate concentration unconcealed that the reaction was looking on KIO4 as Associate in nursing oxidizer. The best absorbance was earned once the concentration of KIO4 was (1×10^{-3}M-9×10^{-3}) higher than this worth a decrease within the absorbance reading occurred Figure (3). Therefore, 1mL of 3×10^{-3}M was used throughout the following work.

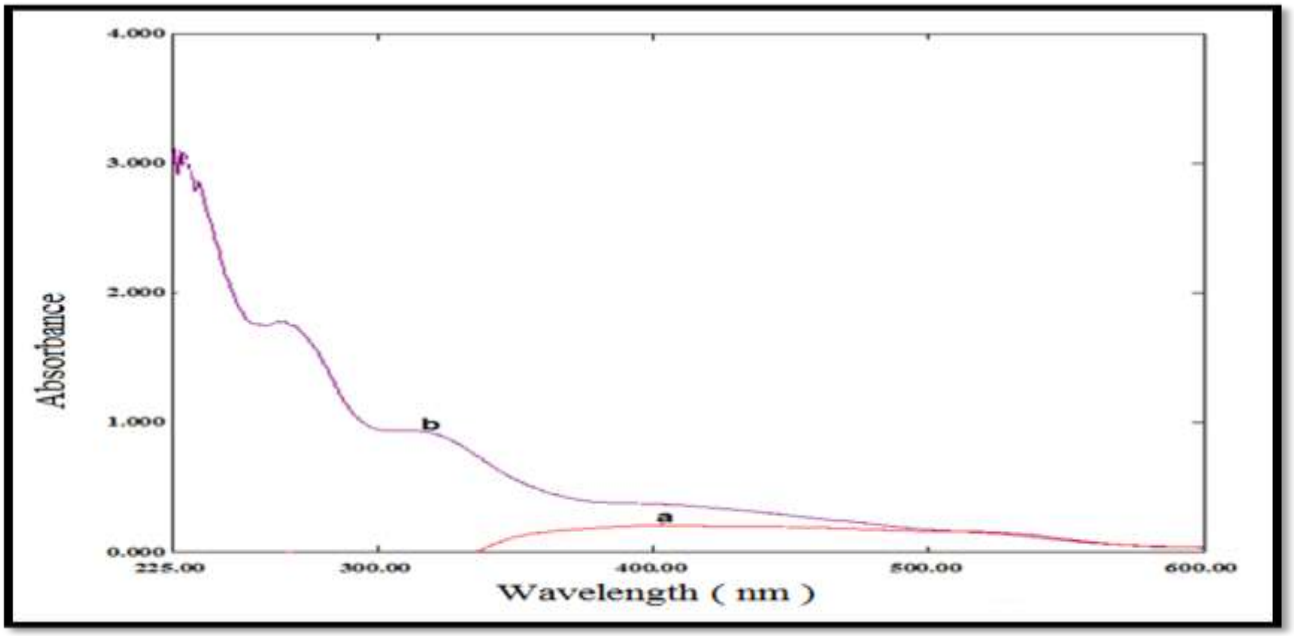


Figure 1: Absorption spectra of: (a) 20 µg.mL-1 (ATN.) against reagent blank, (b) blank solution against solvent under the primary test conditions

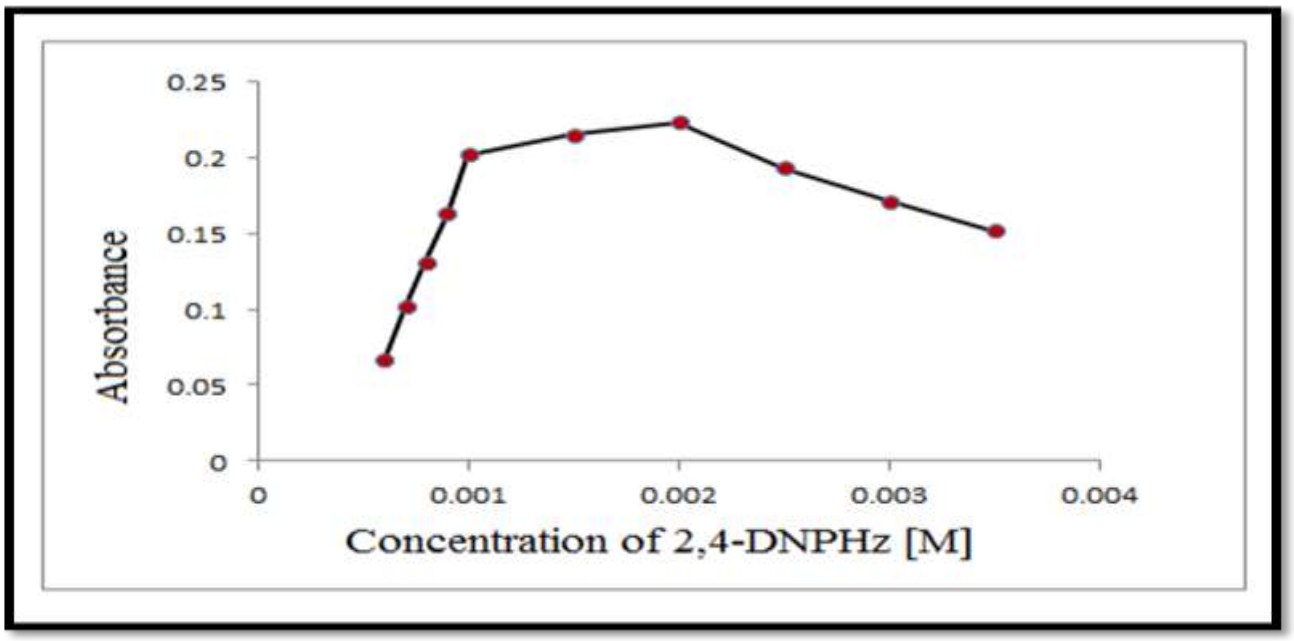


Figure 2: The effect of the concentration of 2, 4-DNPHz on the color development in the determination of 20 µg.mL-1 (ATN)

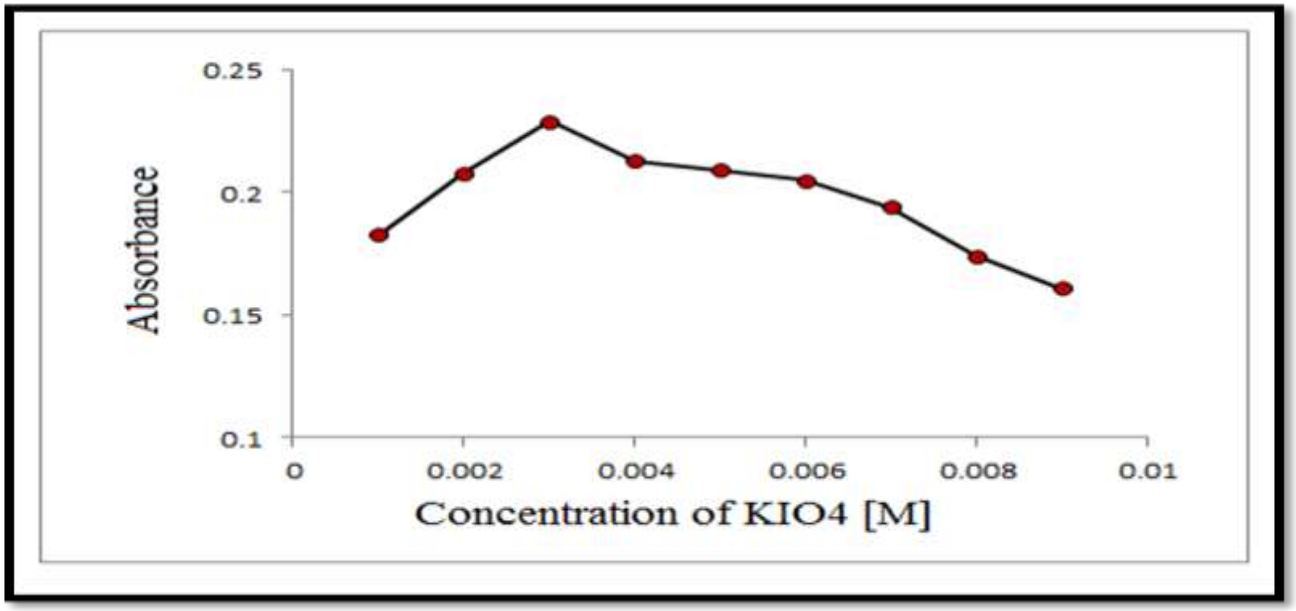


Figure 3: The effect of potassium periodate concentration on the color development in the determination of 20 µg.mL-1(ATN)

Effect of Different Bases

The impact of various a calescent solutions with concentration of 1M on the absorption intensity of the colored dye fashioned was investigated. Four styles of bases namely; hydroxide, hydroxide, soda and ammonia were tested and also the results were listed in Table (1). As are often seen it absolutely was found that hydroxide shows the most absorption intensity of the colored product, so it absolutely was designated for later work.

Table 1: The effect of different bases on coupling reaction

Base (1M)	Absorbance
NaOH	**0.229**
KOH	0.181
Na2CO3	0.003
NH4OH	0.127

Effect of Sodium Hydroxide Concentration

The result of hydroxide concentration on the measured absorbance of the shaped colored product was investigated by victimization 1mL of various concentrations of NaOH resolution ranged between (0.1-3.0) M.

The results square measure bestowed in Figure (4) that reveal that the addition of 1mL of 1M NaOH exhibited a more robust absorbance. On top of this concentration the absorbance worth belittled. Therefore, 1mL of 1M NaOH was employed in all resulting experiments.

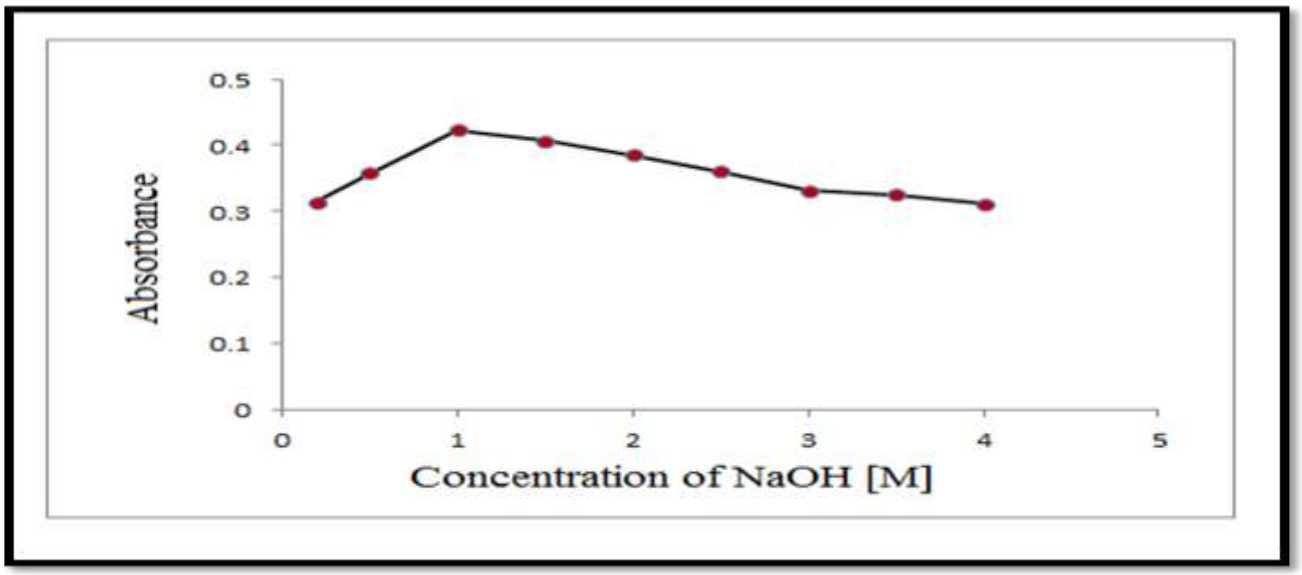


Figure 4: The effect of sodium hydroxide concentration on the color development in the determination of 20 µg.mL-1 (ATN.)

Effect of Coupling Reaction Time

The optimum time for the reaction between (ATN.) and 2,4-DNPHz was studied at a set concentration of (ATN.). 20µg.mL^{-1} reacted with 2,4-DNPHz and metallic element periodate in alkalescent medium. Absorbance values were recorded at completely different intervals starting from immediate measure to a waiting amount of 5min. The aerobic coupling reaction is completed in 5 min as shown in Table (2).

Table 2: The effect of coupling reaction time

Time (min)	Absorbance
0	0.173
1	0.198
2	0.219
4	0.223
5	0.228
7	0.215
10	0.191
12	0.183
15	0.175

Effect of Reagents Mixing Order

Effect of various orders of elements addition on chromogenic formation was investigated by dynamic the order of addition of reactants fourfold as shown in Table (3). From the results shown, it's obvious that mix order best was counseled because it resulted in getting a most absorbance and therefore was followed within the sequent experiments.

Table 3: Variation of absorbance with change of reactants addition order in the determination of 20 µg.mL-1 (ATN)

NO	Sequence	Absorbance
1	R+O+D+B	0.241
2	R+D+O+B	0.225
3	D+R+O+B	0.232
4	D+B+R+O	0.006

R: reagent O: oxidizing agent D: drug B: base

The Stability

Stability study of the colored product shaped upon reaction of drug answer with 2,4-DNPHz was disbursed by mensuration its absorbance at completely different time intervals 20min was chosen as optimum time within the general suggested procedure. The cooler of the answer was nearly stable for a minimum of 60min as shown in Figure (5).

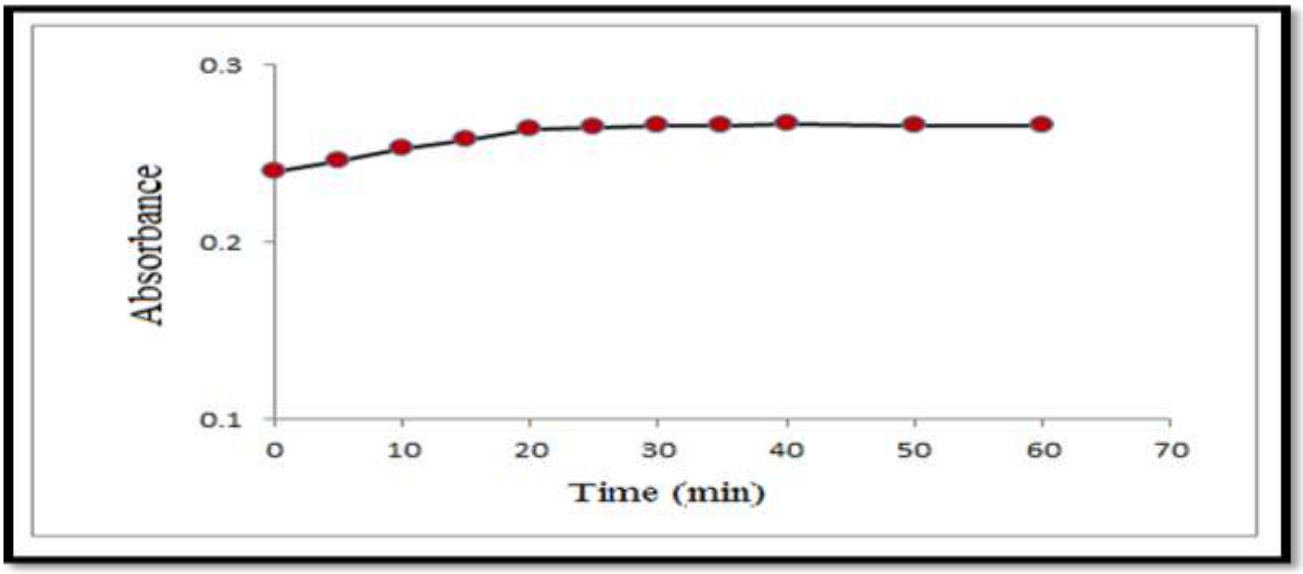


Figure 5: The stability of the colored product with time

Final Absorption Spectra

The spectrum of the yellowish brown product shaped from the treatment of (ATN.) 20µg.mL-1 with 2,4-DNPHz within the presence of metallic element periodate in base-forming medium beneath the optimum conditions was recorded and showed a most absorption at 403nm against the chemical agent blank as shown in Figure (6).

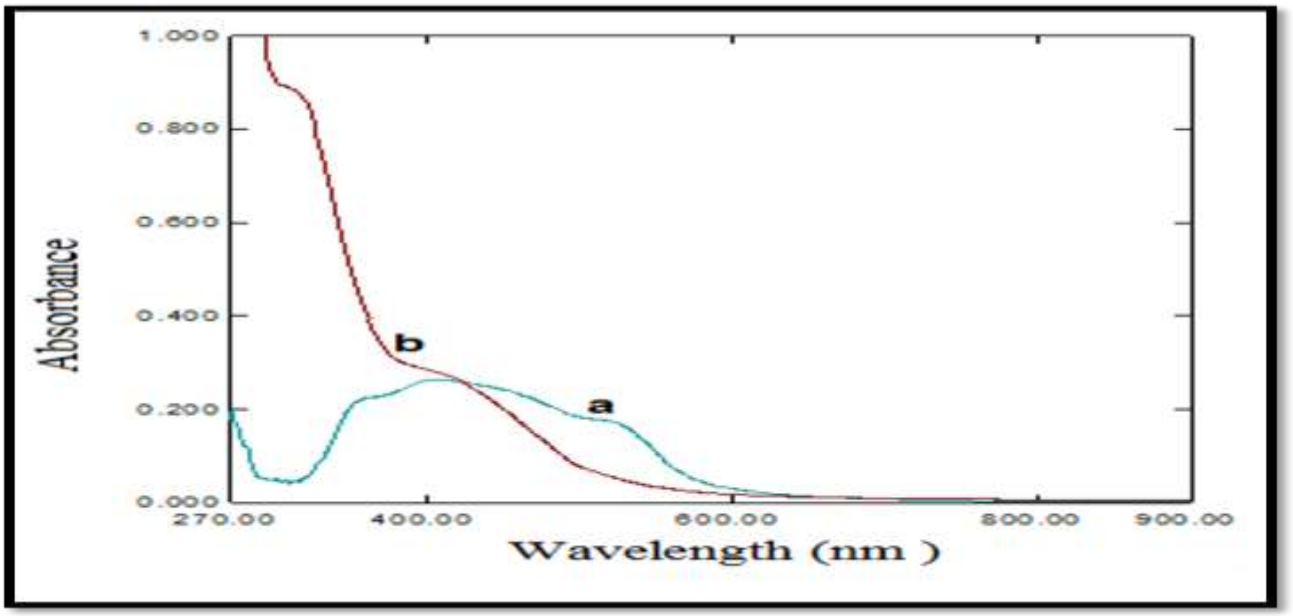


Figure 6: Absorption spectra of: (a) 20 µg.mL-1(ATN.) against reagent blank, (b) blank solution against solvent under the optimum conditions

Calibration Curve and Analytical Data

Employing the optimum process, the measured absorbance values at 403 nm versus completely different normal concentrations of (ATN.) were planned to construct a standardization curve.

The dimensionality of the obtained plot of the (ATN.) was within the concentration vary of (1- 30) µg.mL-1 as shown in Figure (7). The applied mathematics treatments of the analytical information square measure summarized in Table (4).

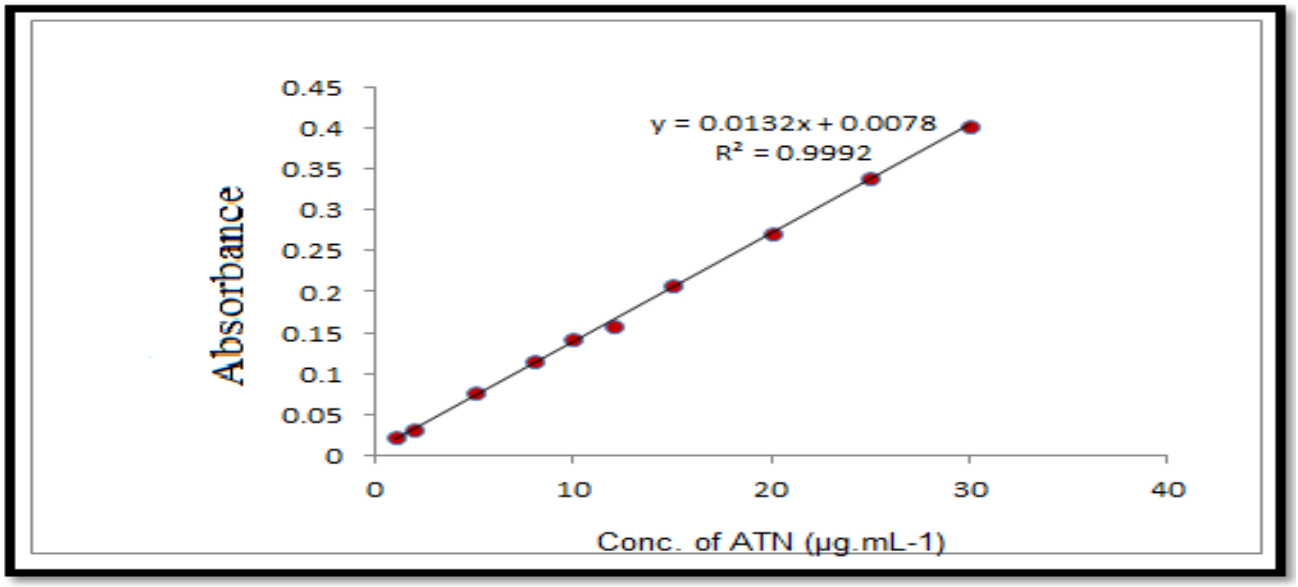


Figure 7: Calibration curve for the determination of (ATN.) under optimum conditions

Table 4: Optical characteristics and statistical data for the determination of (ATN)

Parameter	Value
λ max (nm)	403
Color	Yellow-green
Regression equation	y=0.0132[ATN]+0.0078
Linearity range (µg.mL-1)	0.0132
Calibration sensitivity (mL.µg-1)	1-30
Correlation coefficient (r)	0.9995
Correlation of linearity (R2)	0.9992
Molar absorptivity (L.mol-1.cm-1)	3.5×10^4
Sandal's sensitivity (µg.cm-2)	0.07575
L.O.D. (µg.mL-1)	0.2536
L.O.Q. (µg.mL-1)	0.7686

Structure of the Product

Job's technique [10] and mole quantitative relation technique [11] are employed in the determination of the ratio of the reaction between (ATN) and 2, 4-DNPHz. The obtained results Figures (8) & (9) showed that 1:1 beta-adrenergic blocking agent to 2, 4-DNPHz quantitative relation is obtained. The projected mechanism of the reaction between (ATN.) and 2,4-DNPHz are often delineated in Theme (2).

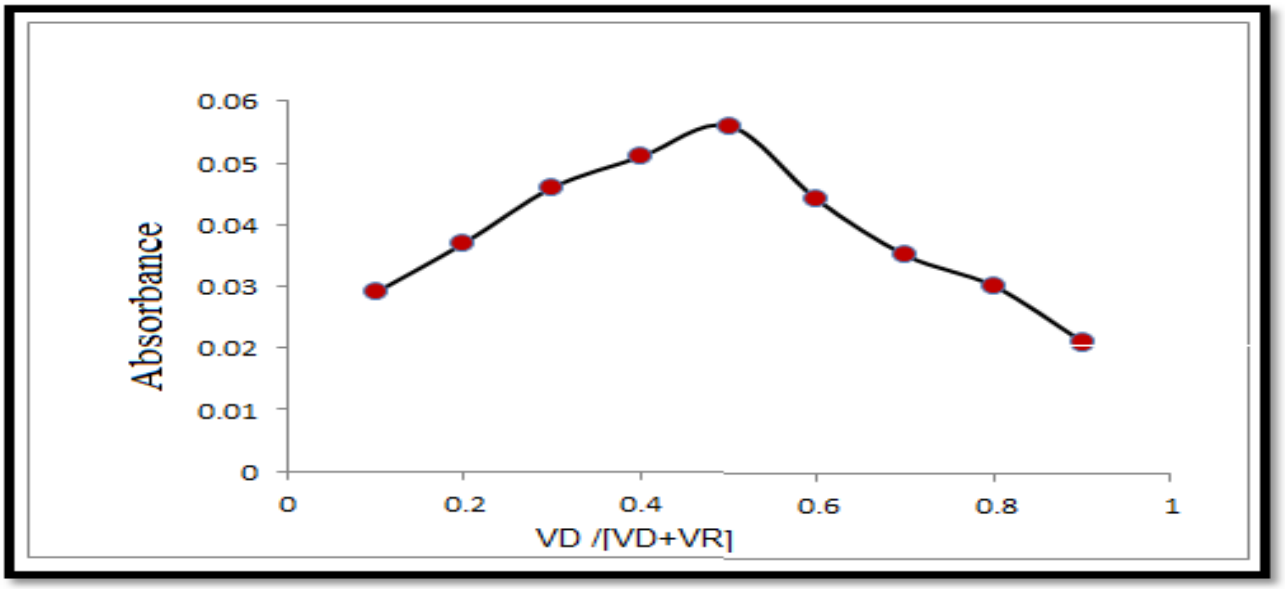


Figure 8: Continuous variation method for the reaction of (ATN.) with 2,4-DNPHz

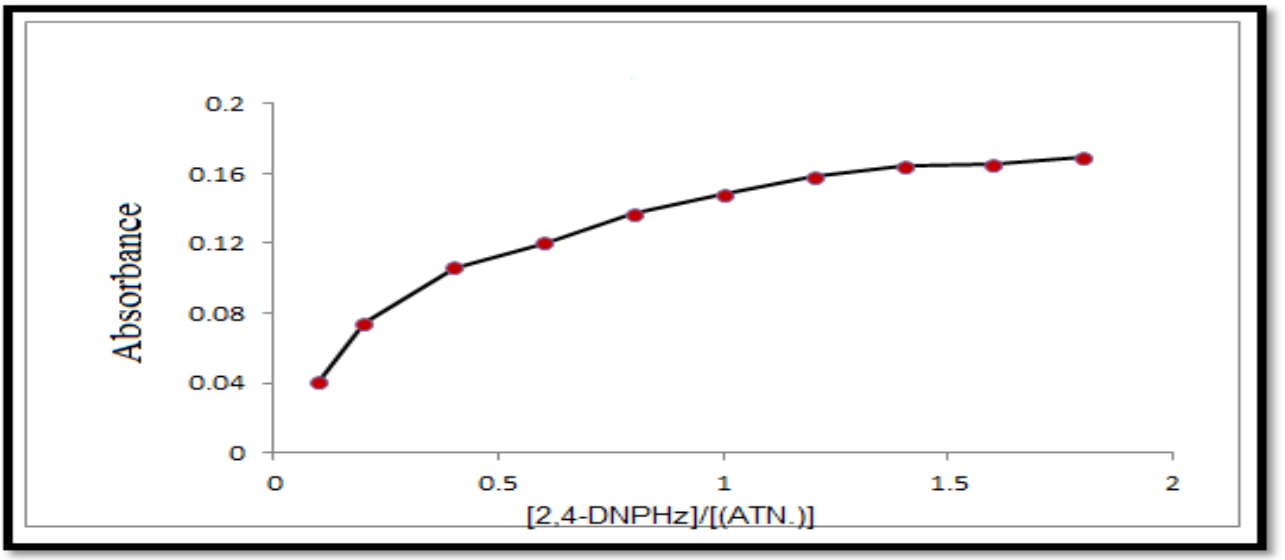
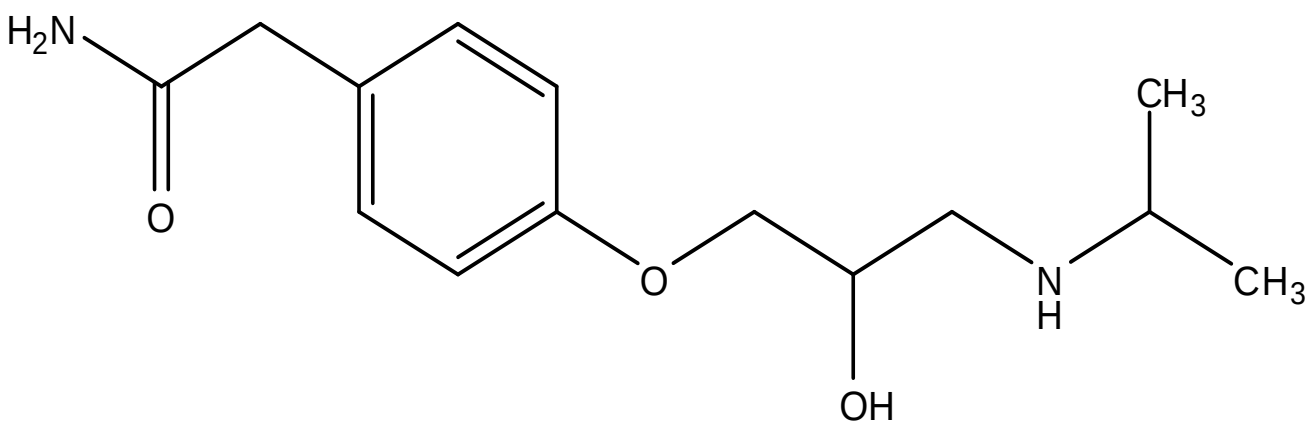
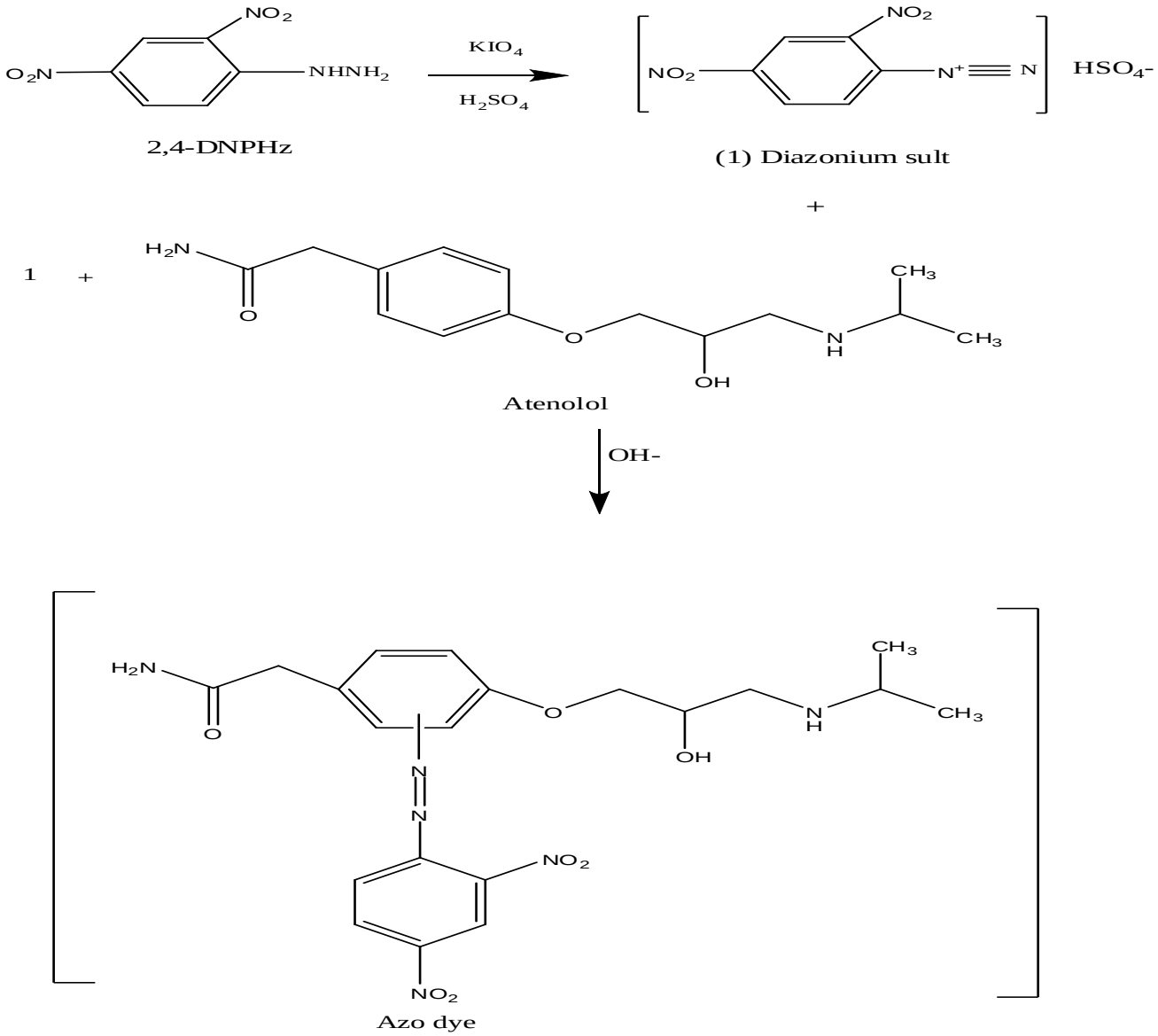


Figure 9: Mole ratio method for the reaction of (ATN.) with 2,4-DNPHz



Scheme 1: The structural formula of Atenolol



Scheme 2: The suggested reaction mechanism between (ATN.) and 2, 4-DNPHz

Comparison of the Methods

Table (5), shows a comparison between the projected methodology which of another literature spectrophotometric ways throughout some measured analytical parameters.

Table 5: Analytical parameters for the analysis of Atenolol by the proposed method comparing to other methods

Methods	Linear range µg.mL-1	Correlation Coefficient (R)	C.V% range	Ref.
Proposed method	1.0 - 30.0	0.9995	0.319-1.45	....
Spectrophotometric	2.0 – 14.0	0.9999	0.337-1.428	28
analysis Flow-injection	0.1 – 20	0.9869	0.17-0.54	29
Spectrophotometric	5.0 – 60	0.9997	0.221-0.693	30
liquid chromatography	4.0 – 12.0	0.9984	0.25 – 1.08	31
HPLC–UV	1.0 – 25.0	0.9994	0.20 – 0.87	32

Precision and Accuracy

The preciseness and accuracy of the projected technique was tested by analyzing 3 replicate samples of (ATN.) in 3 totally different concentration levels (within Beer's law range). The results listed in Table (6) indicate an appropriate accuracy and preciseness of the strategy.

Table 6: Evaluation of the accuracy and precision of the proposed method for the determination of (ATN)

Conc. of (CRN.) µg.mL-1		Er%	C.V%
Taken	Found*		
8	8.05	0.625	1.32
15	15.29	1.93	0.54
20	20.04	0.20	2.57

Interference Study

The extent of busy by some excipients which regularly attended pharmaceutical preparations was studies by measurement the absorbance of answer containing twenty μg.mL^{-1} of (ATN.) and 1000μg.mL^{-1} of excipient. The leads to Table (7) show that the studied excipients don't interfere within the determination of (ATN.) in its indefinite quantity forms.

Table 7: Recovery values for 20 μg.mL-1 of (ATN.) in the presence of 1000 μg.mL-1 of different excipients

Excipients		Carbamazepine Conc.		Recovery Name (%)
Name	Conc. (μg.mL-1)	Taken (μg.mL-1)	Found (μg.m-1)	
Lactose	1000	20.000	19.863	99.315
Glucose			20.015	100.075
Sucrose			19.787	98.935
Starch			20.242	101.21

Application in Pharmaceutical Preparation

In order to demonstrate the pertinence of the planned methodology for the determination of (ATN.), the tactic was applied to two styles of pharmaceutical formulations (tablets) from completely different producing sources containing (ATN.). The results of the applying were satisfactory as shown in Table (8).

Table 8: Determination of Atenolol in pharmaceutical formulations (tablets) by the proposed method

Sample	Weight Found* (mg)	Concentration (μg.mL-1)		Recovery %	C.V %
		Taken	Found*		
TENORMIN 50 mg	49.93	10	9.93	99.30	0.755
	51.02	20	19.91	99.55	0.431
TENORMIN 100 mg	99.95	10	9.96	99.60	0.863
	100.04	20	19.94	99.70	0.351
VASCOTEN100 mg	100.24	10	9.98	99.80	0.460
	100.43	20	20.01	100.05	0.659

*Average of three measurements

Conclusions

The reagents utilized within the projected methodology area unit promptly out there, low cost and also the procedures don't involve any vital reaction conditions. Moreover, the tactic is free from interference by excipients. The wide relevance of the new procedure for routine internal control was well established by the assay of carbamazepine in pure type and in pharmaceutical preparations.

References

1. Paterson JW (1982) Citation classic-the pharmacodynamics and metabolism of propranolol in man. Current contents/clinical practice, 20.
2. Reynolds JEF (1982) Ed. Martindale; Extra Pharmacopoeia. 28th Ed. The Pharmaceutical Press: London, 13-37.
3. The Indian Pharmacopoeia. 4th Ed (1996) The controller of publications, the ministry of health and family welfare, Government of India, New Delhi, 72.
4. The British Pharmacopoeia (1988) I. Her Majesty's Stationery Office, London, 49.
5. El-Gindy A, Emara S, Mostafa A (2005) HPLC and chemometricassisted spectrophotometric methods for simultaneous determination of atenolol, amiloride hydrochloride and chlorthalidone. IL Farmaco, 60: 269-78.
6. Elgawish MS, Mostafa SM, Elhanan wane AA (2011) Simple and rapid HPLC method for simultaneous determination of atenolol and chlorthalidone in spiked human plasma. Saudi. Pharm. J., 19: 43-9.
7. El-Gindy A, Sallam S, Abdel-Salam RA (2008) HPLC method for the simultaneous determination of atenolol and chlorthalidone in human breast milk. J. Sep. Sci., 31: 677-82.
8. Spanakis M, Niopas I (2013) Determination of atenolol in human plasma by HPLC with fluorescence detection: validation and application in a pharmacokinetic study. J Chromatograph Sci., 51: 128-32.
9. Neelima MS, Gandhi BM, Raju VB, Sumanth KS, Srinivas K, Mounika P, et al (2016) Development and validation of stability indicating a reverse phase high-performance liquid chromatography

method for simultaneous estimation of atenolol, hydrochlorothiazide and losartan in bulk and pharmaceutical dosage form. Asian J. Pharm. Clin Res., 9: 118-24.

10. Yilmaz B (2010) Determination of atenolol in pharmaceutical preparations by gas chromatography with flame ionisation and mass spectrometric detection. Anal Lett., 43: 2311-7.
11. Kannappan V, Mannemala SS (2016) Simultaneous enantioseparation and purity determination of chiral switches of amlodipine and atenolol by liquid chromatography. J. Pharm. Biomed. Anal., 120: 221-7.
12. Lawson G, Cocks E, Tanna S (2012) Quantitative determination of atenolol in dried blood spot samples by LC-HRMS: a potential method for assessing medication adherence. J Chromatogr. B., 897: 72-9.
13. Sartori ER, Medeiros RA, Rocha-Filho RC, Fatibello-Filho O (2010) Square-wave voltammetric determination of propranolol and atenolol in pharmaceuticals using a boron-doped diamond electrode. Talanta., 81: 1418-24.
14. Goyal RN, Singh SP (2006) Voltammetric determination of atenolol at C60-modified glassy carbon electrodes. Talanta, 69: 932-7.
15. Goyal RN, Gupta VK, Oyama M, Bachheti N (2006) Differential pulse voltammetric determination of atenolol in pharmaceutical formulations and urine using nanogold modified indium tin oxide electrode. Electrochem Commun., 8: 65-70.
16. Zzam KA, Elbashir AA, Elbashir MA, Saad B, Hamid SA (2009) Simultaneous determination of atenolol and chlorthalidone in pharmaceutical preparations by capillary-zone electrophoresis. Anal. Lett., 42: 1458-70.
17. Xu L, Guo Q, Yu H, Huang J, You T (2012) Simultaneous determination of three ß-blockers at a carbon nanofiber paste electrode by capillary electrophoresis coupled with amperometric detection. Talanta, 97: 462-7.
18. Wang Y, Wu Q, Cheng M, Cai C (2011) Determination of ß-blockers in pharmaceutical and human urine by capillary electrophoresis with electro chemiluminescence detection and studies on the pharmacokinetics. J. Chromatogr. B, 879: 871-7.
19. Al-Arfaj NA, Al-Abdul Kareem EA, Aly FA (2009) Determination of enalapril maleate and atenolol in their pharmaceutical products and in biological fluids by flow-injection chemiluminescence. Luminescence, 24: 422-8.
20. Basan H, Yarımkaya S (2014) A novel solid-phase extraction–Spectrofluorimetric method for the direct determination of atenolol in human urine. Luminescence, 29: 225-9.
21. Thomas A, Patankar M, Deshmukh KR, Kothapalli L, Jangam S, Patankar PR, et al (2007) Simultaneous spectrophotometric estimation of losartan potassium and atenolol in bulk and two component formulation. Asian J. Chem., 19: 3721-6.
22. Basavaiah K, Chandrashekar U, Nagegowda P (2006) Titrimetric, spectrophotometric and kinetic methods for the assay of atenolol using bromated-bromide and methyl orange. J. Serb. Chem. Soc., 71: 553-63
23. Prashanth KN, Basavaiah K (2012) Sensitive spectrophotometric determination of atenolol in pharmaceutical formulations using the bromate-bromide mixture as an Eco-friendly brominating agent. J. Anal. Methods Chem., 2012: 1-12.
24. Al-Ghannam SM (2006) A simple spectrophotometric method for the determination of ß-blockers in dosage forms. J. Pharm. Biomed. Anal., 40: 151-6.
25. Prashanth ken, Basavaiah K, Abdulrahman SAM, Rajendraprasad N, Vinay BK (2012) Application of bromated-bromide mixture as a green brominating agent for the spectrophotometric determination of atenolol in pharmaceuticals. Chem. Ind. Chem. End Q, 18: 43-52.
26. Prashanth KN, Basavaiah K (2012) Simple, sensitive and selective spectrophotometric methods for the determination of atenolol in pharmaceuticals through charge transfer complex formation reaction. Acta Poloniae Pharm., 69: 213-23.
27. Akram M (2017) El-didamony1,†, Moftah A. Moustafa2, Direct Spectrophotometric Determination of Atenolol and Timolol

Antihypertensive Drugs , International Journal of Pharmacy and Pharmaceutical Sciences, 9: 3. A1.

28. Eisam M Ali, Naghm S Aleiwadi, Ealaa Alnizam (2013) alkimutalqii lilwaminal-tariqat kimutalqit-hqin jaryaniun mustamirun litaqdir aaltynwlwl muetamadaan hydrwksyd alsswdywm-biruksyd alhydrwjyn , majalat jamieat alnahrayn, almajalid 61 aledd) 4) kanun yatawaqaful ,13: 1.
29. Hayam Mahmoud Lotfy ⇑, Maha A Hegazy, Mammoth R Rezk, Yasmin Rostom Omran (2014) Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy, Analytical Chemistry Department, Faculty of Pharmacy, Cairo University, Kasr El-Aini Street, 11562 Cairo, Egypt, Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy, 126: 197-207.
30. KG Baheti*, N Shah, S Shaikh (2012) Ion-Pairing Reverse-Phase High Performance Liquid Chromatography Method for Simultaneous Estimation of Atenolol and Indapamide in Bulk and Combined Dosage Form, Indian Journal of Pharmaceutical Sciences, May-June.
31. Mohan Gandhi Bonthu (2016) Development and Validation of Stability Indicating RP-HPLC Method for Simultaneous Estimation of Atenolol, Hrdrochlorothiazide and Losartan in Bulk and Pharmaceutical Dosage Form, Asian Journal of Pharmaceutical and Clinical Research, 9(2):118-124, January 2016 *with* 365 Reads.

