RESEARCH ARTICLE

Immediate Real-time Estimation of a Combination of Drugs Containing Amino Groups with Ultraviolet Spectroscopy

Imad T. Hanoon[1]*, Mumin F. H. Al-Samarrai[1], Mahmood Z. Lafta[2]

[1]*University of Samarra, Education College, Samarra, Iraq*
[2]*Salah al-Din Education Directorate- Samarra Education Department, Iraq*

Received: 13th February, 2022; Revised: 27th February, 2022; Accepted: 07th March, 2022; Available Online: 25th March, 2022

ABSTRACT

The drugs olanzapine and ephedrine hydrochloride have been estimated in real-time by developing a method that avoids the pre-separation of the two drugs if they are present together, making the method accurate, sensitive, and simple. The method is the derivative of spectral ratio method, in which the absorption spectrum of the mixture of the two drugs is divided by the absorption spectrum of one of the two drugs, which is calculated as an overlap. The outcome of the process is the absorption spectrum of the other drug that is required to be estimated. Then it is derived as the first derivative was used for the absorption spectrum of olanzapine and ephedrine hydrochloride. The results of this method showed compliance with Beer's law, as the range of concentrations ranged from (15–30) to (5–28) $mg.mL^{-1}$, The results were accurate and well-accepted, as they ranged between RSD% (0.00005–0.00808) and R.E% (0.00057–0.02189) for the two drugs, respectively, in this way. The proposed method has been successfully applied in estimating the two drugs in some pharmaceutical firms.

Keywords: Amino Groups, Drugs combination, Derivative of Spectral ratio.

International Journal of Drug Delivery Technology (2022); DOI: 10.25258/ijddt.12.1.42

How to cite this article: Hanoon IT, Hamad Al-Samarrai MF, Lafta MZ. Immediate Real-time Estimation of a Combination of Drugs Containing Amino Groups with UV spectroscopy. International Journal of Drug Delivery Technology. 2022;12(1):225-230.

Source of support: Nil.

Conflict of interest: None

INTRODUCTION

Olanzapine is scientifically called according to the IUPAC system 2-methyl-4- (4-methyl-1-piperazinyl) -10H-thieno (2,3-b) (1,5) benzodiazepine, and it is one of the drugs used to treat schizophrenia and bipolar disorder. It blocks dopamine D1-D5 receptors and serotonin.[1-5] Figure 1 shows the chemical structure of the drug.

As for ephedrine, it is considered one of the drugs that treat bronchoconstriction and is also used in slimming for those suffering from obesity, and this drug is scientifically called [1,2,5-7] (methylamino) -1-phenylpropan-1-ol hydrochloride (-2), and Figure 2 shows the chemical structure of the drug.

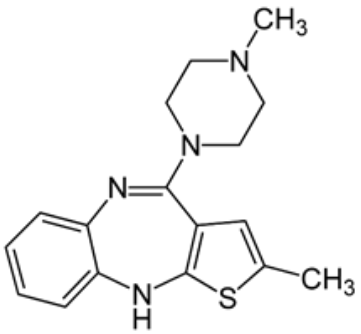

Figure 1: The chemical structure of olanzapine

Although there are many analytical methods used to quantify the two drugs accurately and with good selectivity, using, for example, high-performance liquid chromatography for drug estimation,[8-11] and the use of color interactions to increase the chromophore area.[1,2,12,13] Electrical methods were also used in the quantification of drugs,[14] and the derivative of the spectrum was used to decode the interactions and Calculate the amount of drugs,[2,5,15] and the electro-migration technique was also used to estimate the two drugs.[16]

The aim of this research is to estimate the two drugs Olanzapine and Ephedrine by a method (derivative of spectral ratio) if these two drugs are present together without the need to separate them, which gives the method ease and speed in the estimation.

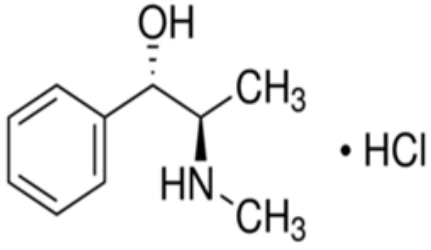

Figure 2: The chemical structure of ephedrine

**Author for Correspondence: dr.imadtarek@gmail.com*

PRACTICAL PART

Devices

- Shimadzu's dual-beam UV-vis Spectrophotometer, model 1800 of Japanese origin, has quartz cells 1 cm wide for all measurements.
- Sartorius BL 210 S sensor scale from Germany, in addition to Dell-Latitude 6430u computer and hp-1100 laser printer to record spectra.

Within the wavelength of 190–350 nm, the absorption spectra of the two drugs and a combination of both were recorded, and the scan speed was medium with the sampling interval is 0.1 nm, the slit width is 2 nm.

Solutions

The standard solutions materials used in work were obtained from the State Company for Medicines and Medical Devices (SDI), Samarra, Iraq.

Standard Solutions

A 250 $mg.mL^{-1}$ was prepared by dissolving 0.025 g of ephedrine hydrochloride and olanzapine separately in a volume of ethyl alcohol and then completed volume to the limit of the mark for the same solvent in a 100 ml volumetric flask.

Pharmaceutical Solutions

In this step, a solution with a concentration of 100 $mg.mL^{-1}$ was prepared from each pharmaceutical preparation that was worked on, after taking the average weight of a pill or the volume of an ampoule and then dissolving it with ethyl alcohol using a 50 mL volumetric flask, and from the pharmaceutical preparations that were used:

- Ephidin 25 mg. ampoule (POPLAR Bangladesh).
- Ephedrine HCl 8 mg. Tablet (XP laps. Canada).
- Olanzapine 5 mg. Tablet (TAJ PHARMA. India).
- ZYPREXA 10 mg. TABLET (Lilly. USA).

Whatman No. 40 filter paper was used to get rid of suspended matter so that the solutions prepared and used were clear and measurable.

METHOD

Single Estimate of Ephedrine and Olanzapine Hydrochloride

Various quantities were transferred to a series of a volumetric flask of 10 mL of both drugs in the range of concentrations (1-500 micrograms) of each drug and were diluted to the mark with ethyl alcohol and these chains consisted of 80 mixture and the reason for choosing this number of mixtures to work in one batch and at the same time and to reduce errors when preparing solutions, so that the work is somewhat fast, and then 40 were selected in accordance with the sensitivity of the device used to measure the spectrum, With the help of calibration curves, the range of concentrations for each drug was determined, as the range of concentrations for both drugs ranged between (4-24 $mg.mL^{-1}$) and the equation of the straight line for ephedrine ($y = 22.705x – 1.958$) and $R^2 = 0.9982$ while the straight-line rate for olanzapine is ($y = 9.2608x – 2.4087$) and $R^2 = 0.9936$.

By recording the absorption spectrum, the wavelengths were determined at 214 nm for ephedrine, and the wavelengths of olanzapine were 227.8 and 273.9 nm.[5] Also, the zero spectrum was used in the immediate real-time estimation according to the method mentioned below.

A real-time estimate of a combination of ephedrine hydrochloride and olanzapine

Derivative of Spectral Ratio Method[17]

The work in this way is based on three steps; the first includes dividing the absorption spectrum of the two drugs mixture by the absorption spectrum of one of the two drugs, which is calculated as overlap and obtaining the absorption spectrum of the drug to be analyzed as shown in the following equation:

$$Abs.spc1 = Abs.spc.mix / Abs.spc2$$

As for the second step, it is opposite to the first, as the overlapping spectrum is to be analyzed and the other spectrum is the overlapping, as shown in the following equation:

$$Abs.spc2 = Abs.spc.mix / Abs.spc1$$

The third step is implementing the derivation process for all the spectra obtained from the division process and each drug separately, and then the required calculations are performed. As:

Abs.spc.mix: (Abs.spc1 + Abs.spc2): The absorption spectrum that results from mixing the two drugs.

Abs.spc1: The spectrum of absorption of ephedrine.

Abs.spc2: The absorption spectrum of olanzapine.

To perform these 3 stages, a series of 10 mL volumetric flasks are taken following the method adopted above. In each series, different quantities of the ephedrine drug to be assessed are placed (1-500 micrograms).

Then, a known and specific amount of olanzapine is added to each series. These quantities for each series are between (1-500 micrograms); when analyzing olanzapine, a series of 10 mL volumetric flasks are also taken. Each series contains different quantities of olanzapine (1-500 micrograms), and then a known and specific amount of ephedrine is added to each series, and these quantities for each series are between (1–500 μg), the volume is then supplemented to the mark with ethyl alcohol.

Absorption spectra were recorded against the synthetic solution (blank, ethyl alcohol only); With the help of the values of the derivative of spectral ratio, the concentrations of each drug are determined.

RESULTS AND DISCUSSION

Absorption Spectra

As in Figure 3, the absorption spectra of ephedrine and olanzapine and their mixtures are recorded, and (a) the absorption spectrum of ephedrine (17 $mg.mL^{-1}$) shows the highest absorbance at the wavelength of 214 nm, (b) the absorption spectrum of olanzapine (15 $mg.mL^{-1}$) and showed two wavelengths at 227.8 and 273.9 nm, (c) the absorption

spectrum of the mixture of the two drugs at a concentration of (20 mg.mL^{-1}) for each drug.

We note from Figure 3 that there is a very large overlap between the two drugs if they exist together, and therefore it is extremely difficult to estimate it by normal methods, so in this work, a spectral method was developed to estimate such mixtures that the overlap between them is large at the lowest cost and without the need to separate them and which need chemicals And time.

Derivative of Spectral Ratio Method

In the method adopted with this work, the absorption spectrum of the two drugs mixture was divided into the absorption spectrum of one of the two drugs, which is calculated as an overlap, and the result of the process is the absorption spectrum of the other drug that is required to be analyzed, and then the derivation of the resulting spectrum is made. when estimating the drug ephedrine in the mixture, the concentration of the mixture was divided by the concentration of the drug olanzapine, which is calculated as interferon. It was found through the method that the best concentration of the crossover drug olanzapine is (12 mg.mL^{-1}).

Figure 4 shows the spectrum of absorption of the drug ephedrine resulting from the division process and the linearity range of (15-30 mg.mL^{-1}). These concentrations were derived using the first derivative, and Figure 5 shows the spectrum of the drug derivative, taking advantage of the height of the peak at the baseline. The drug was estimated at wavelengths (246, 268, 292.6, and 316) nanometers and a peak to a peak at the wavelength (246-268) nanometers and the area under the beam between the wavelengths (244-250), (258-270), (284-298) and (308-326 nm). In the same way, the drug olanzapine was estimated, as the drug ephedrine was considered interlinked, and the first derivative was also used in the assessment, and the best concentration of ephedrine, which we could divide the mixture on, was (25 mcg.mL^{-1}), and Figure 6 shows the resulting olanzapine absorption spectrum. From the division process, as the linearity ranged between (5-28 mg.mL^{-1}), meaning that it increased compared to the above individual method, depending on the height of the peak at the baseline at the wavelengths (226, 270, 278 and 304) nanometers, the peak to the peak (270-278) nanometers and the area under the beam between the wavelengths (222-236) and (274-288) nanometers, it was estimated the drug, and Figure 7 shows the spectrum of the first derivative of the drug.

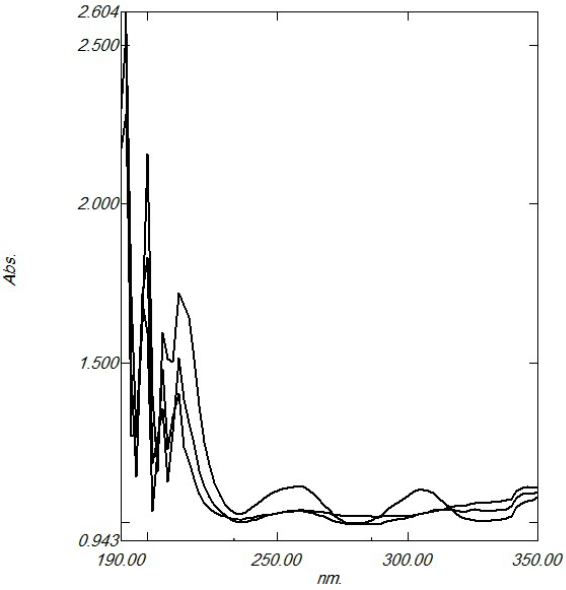

Figure 4: Shows the absorption spectrum of ephedrine at concentrations of (0.5-50 mcg.mL^{-1}) resulting from dividing the mixture's spectrum by the absorption spectrum of olanzapine at a concentration (12 mg.mL^{-1}).

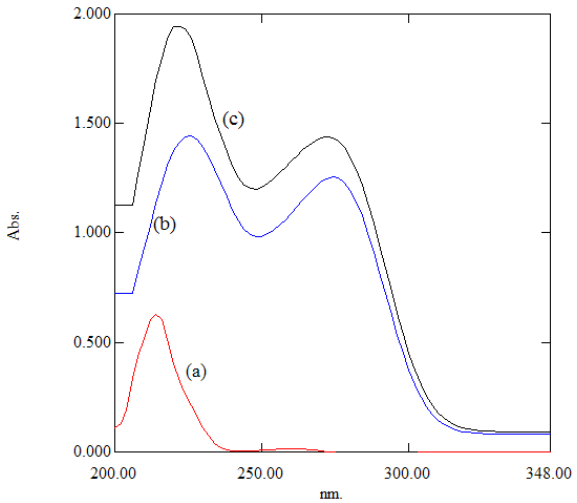

Figure 3: Absorption spectrum (a) ephedrine 17 mg.mL^{-1}, (b) olanzapine 15 mg.mL^{-1}, (c) a mixture of (20 mg.mL^{-1}) for both ephedrine and olanzapine

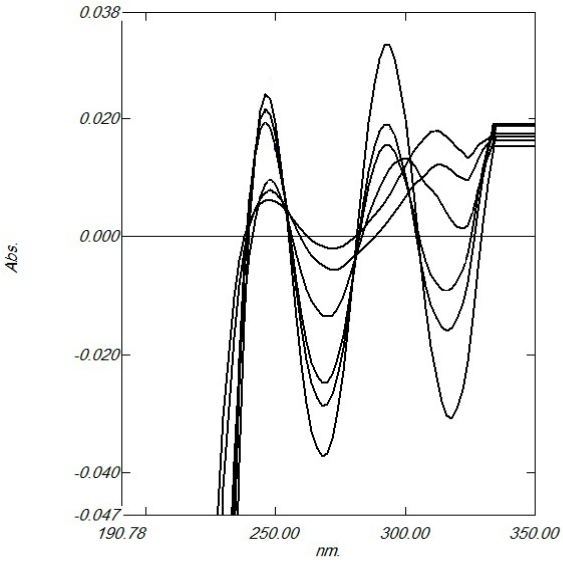

Figure 5: The spectrum of the first derivative shows the spectrum of absorption of the ephedrine drug at concentrations (15-30 mg.mL^{-1}) resulting from the division process.

Calculations and Calibration Curves

By taking advantage of the optimal experimental conditions from the selection of the concentrations that were worked on, and after performing the mathematical calculations and most of the analytical properties, it was found that the linearity of the graphs of the calibration curves was different between the two drugs, as the linearity of the concentrations in the derivative method of olanzapine and ephedrine ranged between (5–28 mg^{-1}) and (15–30 $mg.mL^{-1}$), and the correlation coefficient values ranged between (0.9995-0.9947) and the detection limits values between (1.53093-0.29938 $mg.mL^{-1}$) for the two drugs respectively. Table 1 shows these statistics.

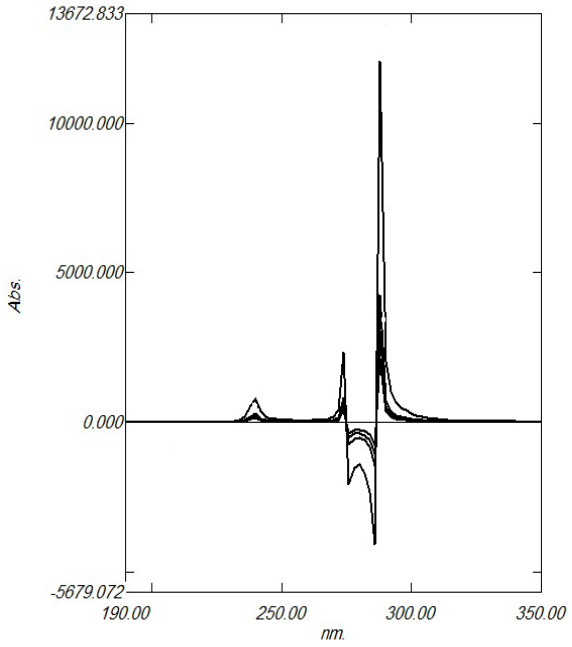

Figure 6: Shows the absorption spectrum of olanzapine at concentrations (0.5–50 $mg.mL^{-1}$) resulting from dividing the mixture's spectrum by the absorption spectrum of ephedrine at a concentration of (25 $mg.mL^{-1}$).

Accuracy and Compatibility

After determining the best conditions and calculating the accuracy and compatibility of the proposed method (for the derived spectral ratio technique), It was found that the percentage of relative error value ranged between (0.00057-0.02189), while the percentage of the relative standard deviation was between (0.00005-0.00808), for olanzapine. And ephedrine, respectively, and for several different

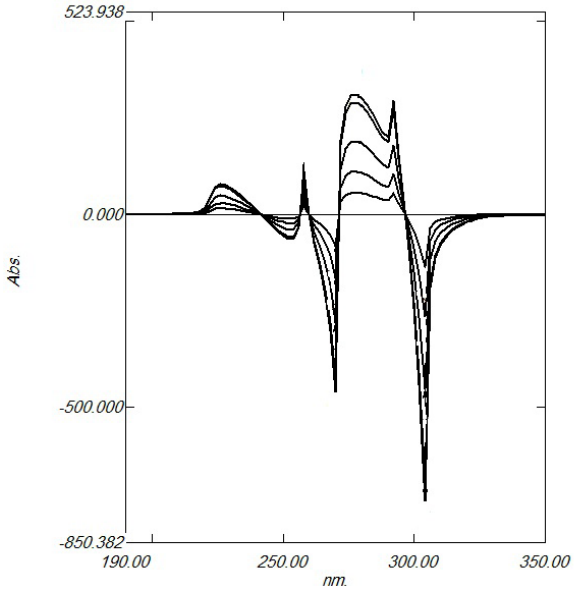

Figure 7: The first derivative spectrum shows the absorption spectrum of olanzapine in concentrations (5-28 $mg.mL^{-1}$) resulting from the division process.

Table 1: Results of olanzapine and ephedrine drug analysis using derivative of spectral ratio method.

Compound	*Order of derivative*	*Mode of calculation*	*λ (nm)*	*Regression equation*	R^2	*D. L.* $\mu g.mL^{-1}$	*L.O.Q* $\mu g.mL^{-1}$
Olanzapine drug	First	Peak to base-line	226	y = 2.8471x - 0.9278	0.997	1.42174	4.73913
			270	y = -17.78x + 21.103	0.9961	1.56234	5.20780
			278	y = 11.58x - 14.405	0.9986	0.98895	3.29649
			304	y = -28.06x + 22.989	0.9992	0.78624	2.62080
		Peak to Peak	270–278	y = -5.7571x + 7.3126	0.9973	1.35520	4.51734
		Peak area	222–236	y = 12.905x - 10.451	0.999	0.84442	2.81496
			274–288	y = 16.313x - 12.519	0.9954	1.49568	4.98561
Ephedrine HCl drug	First	Peak- to-base-line	246	y = 0.001x + 0.0051	0.9995	0.45246	1.50822
			268	y = -0.0022x + 0.0092	0.9947	1.50346	5.01154
			292.6	y = 0.0023x - 0.0172	0.9976	1.13012	3.76770
			316	y = -0.0035x + 0.0426	0.9965	1.47550	4.91833
		Peak to Peak	246–268	y = -0.0012x + 0.0131	0.9977	1.11253	3.70843
		Peak area	244–250	y = 0.0008x - 0.0037	0.9868	1.53093	5.10310
			258–270	y = -0.0033x + 0.0103	0.9967	0.74227	2.47423
			284–298	y = 0.009x - 0.0724	0.9994	0.29938	0.99794
			308–326	y = -0.0187x + 0.1867	0.9973	0.65494	2.18314

Table 2: Calculation of the accuracy and compatibility of the results of evaluating olanzapine and ephedrine by the proposed method of work

Compound	*Method of analysis*	*Taken (μg.mL^{-1})*	*Fond * (μg.mL^{-1})*	*Relative error %*	*Relative standard deviation %*
Olanzapine drug	Derivative of ratio spectra method	7	6.850	0.02189	0.00808
		19	19.031	-0.00163	0.00005
Ephedrine drug	Derivative of ratio spectra method	16	15.970	0.00187	0.00217
		28	28.016	-0.00057	0.00614

* Average of three determinations.

Table 3: Estimate of olanzapine and ephedrine in some pharmaceutical firms, according to the proposed method.

Pharmaceutical preparation	*Method of analysis*	*Labeled amount mg/tablet and capsule*	*Found amount mg/tablet and capsule*			
			*Mean value**	*RSD%*	*E%*	*Rec%*
Ephidin 25 mg. ampoule (POPLAR Bangladesh)	Derivative of ratio spectra method	25	24.7627	0.0201	-0.9493	99.0507
Ephedrine HCl 8 mg tablets (XP laps. Canada).	Derivative of ratio spectra method	8	7.9723	0.0070	-0.3458	99.6542
Olanzapine 5 mg. tablet (TAJ PHARMA. India).	Derivative of ratio spectra method	5	5.0989	0.0246	1.9776	101.9776
ZYPREXA 10 mg. TABLET (Lilly, USA).	Derivative of ratio spectra method	10	9.9967	0.0105	-0.0333	99.9667

* Average of three determinations

Table 4: A comparison between the results of the proposed method and the results of other methods.

Drug	*Method*	*R.S.D%*	*R.E%*	*L.O.Q μg.m^{L-1}*	*D.L. μg.m^{L-1}*	*Rec%*	*Ref.*
Olanzapine drug	Batch and flow-injection	<1	……	……	2.17	97.6	18
	Potentiometric	<5	……	……	……	99.68	19
	Derivative of ratio spectra method*	0.0201	0.0218	2.6208	0.7862	101.97	
	Third and Fourth-order derivative	0.5026	-2.3889	2.5788	0.8510	……	5
Ephedrine drug	HILIC-MS/MS	0.95	0.48	……	……	……	20
	Derivative of ratio spectra method *	0.0070	0.0018	0.9979	0.2993	99.654	

* Proposed modalities

ranges of concentrations for both drugs. Table 2 shows these values.

Application of the Method

To determine the degree of effectiveness of the proposed method in the application, the two drugs under study were analyzed in many pharmaceutical forms, and they were well successful. Table 3 shows the results of the estimation.

Comparison of Proposed Methods

By taking advantage of the optimal conditions and after comparing the results of the proposed method with some of the results of other methods, it was found that the method is accurate and sensitive. Thus, the method is one of the important methods in estimating a mixture of olanzapine and ephedrine. Table 4 shows the comparison of the results of the method with the results of other methods.

CONCLUSIONS

When comparing this method with other spectroscopic methods, this method is good due to its good analytical properties.

When comparing the method with other spectral methods, we find that this method is simple, sensitive and of high accuracy. When applying this method, these two drugs were estimated simultaneously in pharmaceutical firms.

REFERENCES

1. Khalaf FA, Sarmad BD, Mumin FA. Spectrophotometric determination of ephedrine-hydrochloride in pure and pharmaceutical preparations based on charge transfer complexation method. International Conference on Recent Trends in Engineering Science and Management. 2015; 5758-66. Available from: http://data.conferenceworld.in/ICRTESM/P5758-5828.pdf.
2. Mumin FA. Spectrophotometric Determination of Ephedrine-HCl and Olanzapine Drugs Using Charge transfer complexes and Derivative spectrum methods. 2014. Available from: College of education, University of Samarra.
3. Moffat AC, Osselton MD, Widdop B, Watts J. Clarke's analysis of drugs and poisons. Vol. 3. Pharmaceutical press London; 2011.
4. Tantawy MA, Hassan NY, Elragehy NA, Abdelkawy M. Simultaneous determination of olanzapine and fluoxetine hydrochloride in capsules by spectrophotometry, TLC-spectrodensy and HPLC. J Adv Res. 2013;4(2):173–80. Available from: doi: 10.1016/j.jare.2012.05.004.
5. Mumin FH, Imad TH, Diana AS, Khalaf FA, Sarmad BD. Using of Third and Fourth Order Derivative Spectra to Simultaneous Determination of Olanzapine and Ephedrine in Its Pure and

Pharmaceutical Formulations. Ibn Al-Haitham Jour. for Pure & Appl. Sci. 2017;30(1), 266-77. Available from: https://jih.uobaghdad.edu.iq/index.php/j/article/view/1085.

6. Ablulmajead MH, Rola, SN, Lubna, FM. The development Ephedrine hydrochloride estimation method using Spectroscopy (UV-Visi). Diyala Journal. 2008; 29, 1- 3.
7. Al-Majthoub MM, Refat MS, Adam AA, Ahmed MA. Spectroscopic and Intermolecular electron-Transfer Mechanism of Ferric (III) Ephedrine Complex as a Model of Drug design. Int J Electrochem Sci. 2013; 8:7053–61. Available from: http://www.electrochemsci.org/papers/vol8/80507053.pdf.
8. Cruz JC, de Faria HD, Figueiredo EC, Queiroz MEC. Restricted access carbon nanotube for microextraction by packed sorbent to determine antipsychotics in plasma samples by high-performance liquid chromatography-tandem mass spectrometry. Anal Bioanal Chem. 2020;412(11):2465–75. Available from: https://link.springer.com/article/10.1007/s00216-020-02464-4.
9. Liu D, An Z, Li P, Chen Y, Zhang R, Liu L, et al. A targeted neurotransmitter quantification and nontargeted metabolic profiling method for pharmacometabolomics analysis of olanzapine by using UPLC-HRMS. RSC Adv. 2020;10(31):18305–14. Available from: DOI: 10.1039/D0RA02406F.
10. ZHANG Y, LING W, ZHANG Y. Determination of Human Plasma Concentration of Paroxetine and Quetiapine by HPLC[J]. Labeled Immunoassays and Clinical Medicine, 2012, 19(5): 293-295. Available from: http://www.chinabjmy.com/EN/
11. Magdy MA, Abdelfatah RM. Green validated HPTLC and HPLC methods for determination of ephedrine hydrochloride and naphazoline nitrate in the presence of methylparaben, in their pure forms and pharmaceutical formulation. JPC–Journal Planar Chromatogr TLC. 2020;33(2):141–8. Available from: https://link.springer.com/article/10.1007/s00764-020-00024-1.
12. Krebs A, Starczewska B, Puzanowska-Tarasiewicz H, Sledz J. Spectrophotometric determination of olanzapine by its oxidation with N-bromosuccinimide and cerium (IV) sulfate. Anal Sci. 2006;22(6):829–33. Available from: DOI https://doi.org/10.2116/analsci.22.829.
13. Ayman A, Zeid AM, Wahba MEK, Yasser E-S. Analysis of clozapine in its tablets using two novel spectrophotometric reactions targeting its tertiary amino group. Spectrochim Acta Part A Mol Biomol Spectrosc. 2020; 238:118447. Available from: doi.org/10.1016/j.saa.2020.118447.
14. Chavada VD, Bhatt NM, Sanyal M, Shrivastav PS. Dual Fluorescence-colorimetric Silver Nanoparticles Based Sensor for Determination of Olanzapine: Analysis in Rat Plasma and Pharmaceuticals. J Fluoresc. 2020;30(4):955–67. Available from: https://link.springer.com/article/10.1007/s10895-020-02568-1.
15. Mayer A, Copp B, Bogun B, Miskelly G. Identification and characterization of chemically masked derivatives of pseudoephedrine, ephedrine, methamphetamine, and MDMA. Drug Test Anal. 2020;12(4):524–37. Available from: doi.org/10.1002/dta.2764.
16. Zayed S, Belal F. Capillary electrophoresis with field-amplified sample stacking for simultaneous determination of indacaterol and glycopyrronium in inhaler capsules: Application to human plasma and urine. Microchem J. 2020; 155:104779. Available from: doi.org/10.1016/j.microc.2020.104779.
17. Imad TH. Simultaneous Determination for Lansoprazole and Simvastatin drugs via Ultraviolet Spectrophotometry,Tikrit Journal of Pure Scienc. 2017; 8(22), 103-11. Available from: https://www.iasj.net/iasj/article/129584.
18. Jasinska A, Nalewajko E. Batch and flow-injection methods for the spectrophotometric determination of olanzapine. Anal Chim Acta. 2004;508(2):165–70. Available from: doi.org/10.1016/j.aca.2003.11.069.
19. Rajendra PN. Use of Sodium Tetraphenyl Boron for Fabrication of Potentiometric Membrane Sensor for the Assay of Olanzapine in Pharmaceuticals and Human Urine, Applied Chemical Engineering 2019; 2(2). Available from: DOI:10.24294/ace.v2i2.179.
20. Barbora T, Petr K. HILIC-MS/MS Method for Analysis of Ephedrine in Internet-available Drugs, Chromatographia, 2106; 80(4), 523-28. Available from: DOI:10.1007/s10337 -016-3170-5.