World Journal of Pharmaceutical research

Volume 3, Issue 1, 126-138. Research Article ISSN 2277 – 7105

SIMPLE SPECTROPHOTOMETRIC METHOD FOR DETERMINATION OF PAROXETINE IN TABLETS USING CHLORANILIC ACID AS CHROMOGENIC REAGENT

Nwodo Ngozi Justina*, Ibezim Akachukwu

Faculty of Pharmaceutical Sciences, Department of Pharmaceutical and Medicinal Chemistry, University of Nigeria, Nsukka

Article Received on
05 October2013
Revised on 11 November 2013,
Accepted on 02December 2013

***Correspondence for Author:**

Dr. Ngozi Justina Nwodo

Faculty of Pharmaceutical Sciences, Department of Pharmaceutical and Medicinal Chemistry, University of Nigeria, Nsukka.

nwodong@hotmail.com

ABSTRACT

Spectrophotometric analysis of Paroxetine-chloranilic acid complex was studied. Stable and instantaneous purple complex was formed on mixing Paroxetine solution with chloranilic acid in dioxin at ambient temperature. The Job's plot of continuous variation method indicated 2:1 stoichiometry for the interaction and the product exhibited maximum absorption band at 540 nm. The complexation attained equilibrium after 10 minutes from time of mixing and was found to be stable over a period of 2 hrs. Beer's law was obeyed over the concentration range of 5 – 40 µg/ml. The assay limits of detection and quantitation were 0.24 and 0.91 µg/ml respectively. The precision of the method and the values of relative standard deviation never exceeded 2%. No interference was observed from the excipients commonly present in dosage form. The proposed method was successfully applied to the analysis of Paroxetine in pure and pharmaceutical dosage forms with good accuracy, precision and reproducibility.

Keywords: Paroxetine, chloranilic acid, spectrophotometry. Charge transfer, n-electron donor.

INTRODUCTION

The term charge transfer complex denotes a certain type of complex which results from interact of an election acceptor and an electron donor with the formation of weak bonds [1]. Charge transfer complex result from a donor acceptor mechanism of Lewis acid-base reaction between two or more different chemical constituents [2]. The formation of electron donor-acceptor complexes can be rapidly assessed for its validity as a simple quantitative analytical method for many drug substances which can act as electron donors. Charge transfer complexation has been employed in the quantitation determination of methldopa [3], hexamethyletetramine [4], flunarizinedihydrochlride [5], amino acids [6], and pyrimethamine [7]. Chloranilic acid and other π-acceptors have been successfully utilized in the determination of a variety of electron donating basic compounds [8 – 11]. Paroxetine: (3S,4R)-3-[(1,3-benzodioxol-5-vloxy)mthyl]-4-(4-fluorophenyl) piperidine (PRX) is a new generation antidepressant drug.

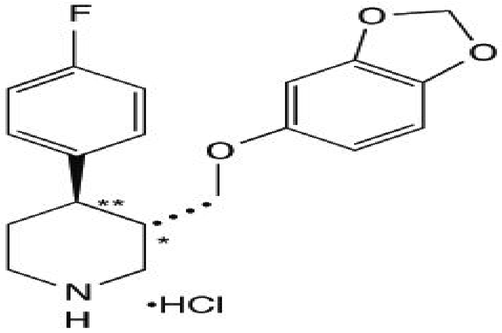


Figure 1. Chemical structure of Paroxetine

It exerts its antidepressant effect through a selective inhibition for the reuptake of the neurotransmitter serotonin by the presynaptic receptor. PRX is comparable to the tricyclic antidepressants in their clinical efficacy, however, PRX is safer and has greater acceptance by the patient [12]. It is also prescribe in the treatment of related disorders, such as obsessive-compulsive disorder, panic fits, social phobia and post-traumatic stress [13]. PRX has been determined in tablets and/or biological fluid by the following methods; voltammetry [14 – 15], densitometry [16 – 17], HPLC [18 – 23], gas chromatography [24 – 26] and capillary electrophoresis [27]. The techniques applied using these methods are tedious and laborious. UV-visible spectrophotometric methods are the instrumental methods of choice commonly used in industrial laboratories because of their simplicity, accuracy, precision and low cost [28 – 30]. This paper describes the application of UV-visible spectroscopy for the quantitative analysis of PRX in pharmaceutical raw and dosage forms.

MATERIALS AND METHODS

Apparatus: UV-Visible spectrophotometer (Jenway 6305) with 1 cm quartz cells was used for all measurements.

Materials and reagent solutions: Paroxetine (GSK Ltd, UK), chloranilic acid (Sigma – Aldrich Co. Ltd., USA). The chloranilic acid was prepared in dioxan at concentration of 0.05 % (w/v). All solvents and other chemicals used throughout this study were of analytical grade.

Pharmaceutical dosage forms: Paroxetine tablets were labelled to contain 20 mg Paroxetine per tablet.

Preparation of standard stock solution: Accurately weighed amount (20 mg) of Paroxetine was transferred into a 100 ml volumetric flask and dissolved in 60 ml of methanol and diluted to volume with methanol to provide a stock solution of 200 µg/ml. The stock solution was diluted with methanol to obtain the suitable working concentrations.

Preparation of sample for analysis: Ten tablets were weighed, finely powdered. An accurately weighed quantity (0.208 g) of the powdered ingredient was transferred into a 10 ml volumetric flask, dissolved in about 6 ml of methanol. The contents of the flask were swirled, sonicated for 20 minutes and then diluted to volume with methanol. The mixture was mixed well, filtered through Whatman paper (No 41) and the first portion of the filtrate was rejected. A measured volume (1 ml) of the filtrate was transferred to a 10 ml volumetric flask and diluted quantitatively with methanol to give a working solution containing 20 µg/ml.

General assay procedure: Aliquots of standard or sample solution were transferred into a 10 ml volumetric flask. Then 1 ml of chloranilic acid (0.05% w/v) was added. The reaction solution were mixed well and allowed to stand at room temperature for 20 minutes. The solutions were completed to volume with dioxan. The absorbance readings were measured at λmax 540 nm against reagent blank treated similarly.

Stoichiometry of the complex species: The Job's method of continuous variation was employed [31]. Equimolar solutions (9.75×10^{-5} M) of Paroxetine and chloranilic acid were prepared. The drug was prepared in methanol and in dioxan for chloranilic acid. Series of 10-ml portions master solutions of the drug and the reagent were made up comprising different complimentary ratios (0:10, 1:9 … 9:1, 10:0 inclusive) in 10-ml volumetric flasks. The reactions were allowed to proceed under the optimum conditions reported under the general assay procedure. The absorbance readings of the resulting solutions were measured at λmax 540 nm against reagent blank treated similarly. The composition of the system was further ascertained by slope ratio method wherein two series of solutions were prepared with a constant concentration of chloranilic acid solution and the other containing varying amounts of Paroxetine and vice versa.

RESULTS AND DISCUSSION

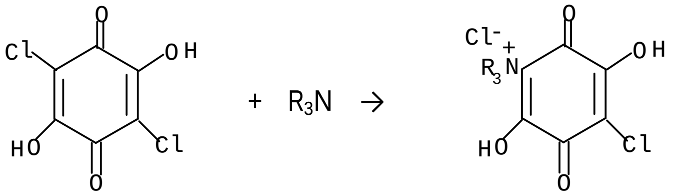
The reaction of chloranilic acid with Paroxetine possessing a lone pair of electrons results in the formation of a charge-transfer complex of the n-π type. The formation of the complex is assumed to occur through a partial ionic bond (D^+A^-).

$D + A \longrightarrow [D^+A^-] \longrightarrow D^+ + A^-$

Donor Acceptor Donor-acceptor Radical anion Complex

In the above reaction, chloranilic acid acts as electron acceptor and Paroxetine (containing tertiary amine) as electron donor. Previous report has suggested the reaction between quinonones (acting as electron acceptor) and tertiary amines to involve quanternization reaction with liberation of chloride. Thus scheme 1 illustrates the reaction between chloranilic acid and tertiary amine, while Figure 2 depicts the spectrum of chloranilic acid and the complex respectively.

Scheme 1:



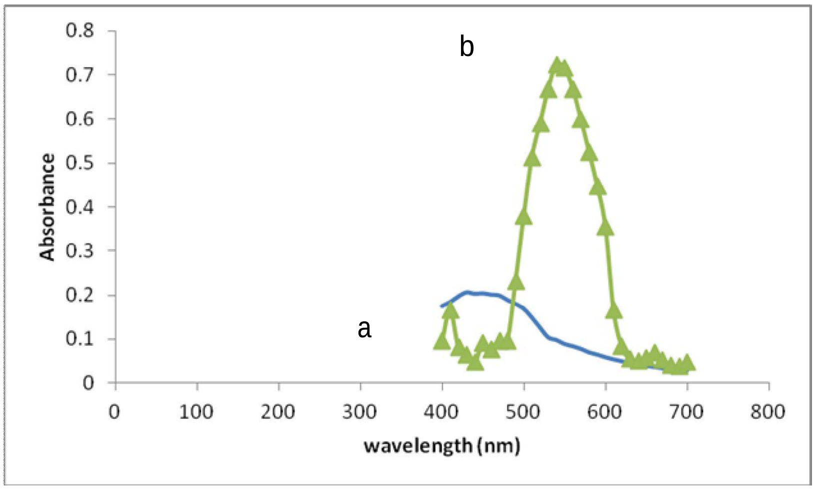


Fig. 2: Absorption spectra of chloranilic acid (a) and Paroxetine-chloranilic acid complex (b)

Optimization of reaction conditions: A series of experiments were performed to establish the optimum experimental conditions for the charge-transfer complexation of Paroxetine with chloranilic acid. The variables affecting the reaction were concentration of chloranilic acid solution, reaction time and diluting solvents. These variables were optimized, by altering each variable in turn while keeping the other constant.

Concentration of chloranilic acid: The effect of chloranilic aicd solution was investigated by carrying out the reaction using 1 ml of chloranilic acid solution of different concentrations varying from 0.01 – 1.0 % (w/v). It was found that the absorbance increased by increasing the concentration of chloranilic acid up to 0.05% w/v. Higher concentrations of chloranilic acid solution had no effect on the values of absorbance readings in all cases.

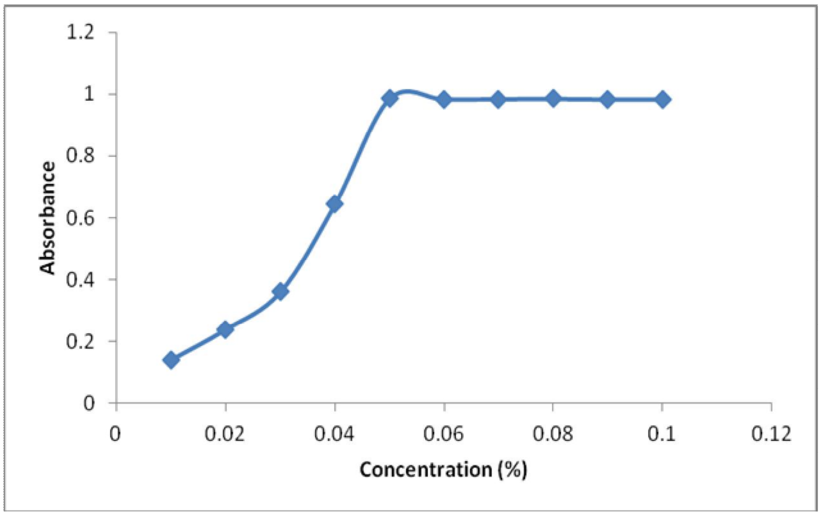


Figure 3: Absorption spectra for optimum concentration of chloranilic acid

Reaction time: The effect of time on the complexation of Paroxetine with chloranilic acid was studied by carrying out the reaction for different periods of time ranging from 2 to 20 minutes. The results revealed that the optimum time (for complete reaction) was 10 minutes and longer time had neither enhancement nor negative effect on the reaction time. For precision considerations, 10 minutes was selected for subsequent studies. Developed colour was stable for more than 1 h, which was considered sufficient time for an analyst to carry out analysis.

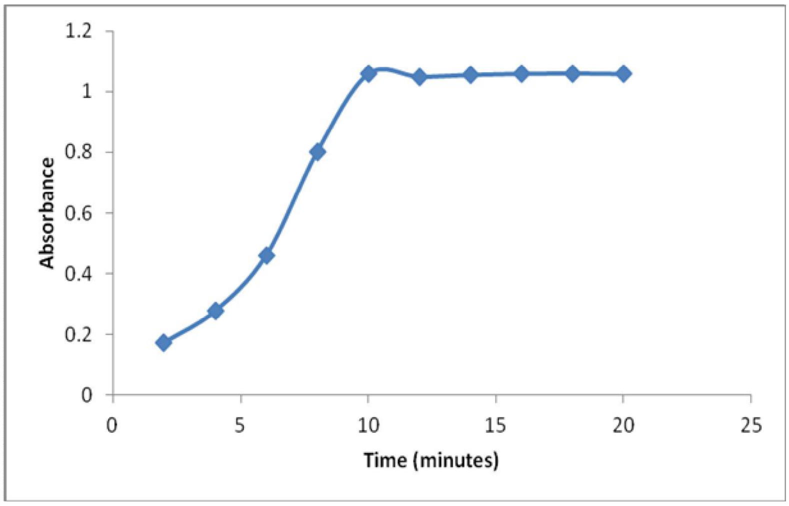


Figure 4: Absorption spectra for optimum reaction time

Diluting solvents: Dilution with different solvents: methanol, ethanol, acetonitrile and dioxan, showed that dioxan was the optimum solvent for dilution as highest absorbance and lowest blank readings were obtained.

Stoichiometry of the reaction: The stoichiometry of the charge-transfer complex was established by the slope molar ratio and Job's method of continuous variation using equimolar solutions of the drug and the reagent. The result obtained is shown in Fig. 3 and indicates that the composition of charge-transfer complex was (2:1) drug to reagent. The center was postulated to be the more basic tertiary amino group in the drug molecule.

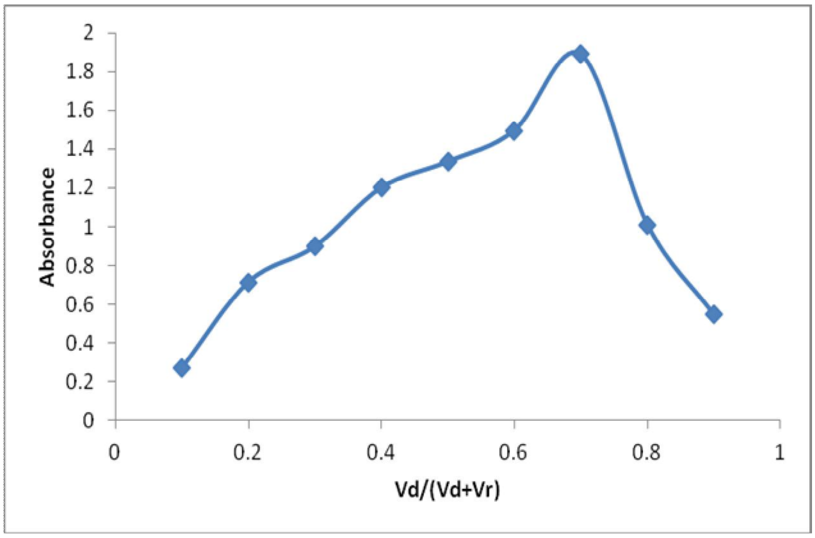


Figure 3: Job's continuous variation plot for the charge – transfer complex, λmax = 540 nm, where, Vd and Vr are the volumes of added drug and reagent respectively.

Validation of the proposed method

Linearity, detection and quantitation limits: Under the specified optimum reaction conditions, the calibration curve for the reaction of Paroxetine with chloranilic acid was constructed by analyzing a series of concentrations of the standard solution of the drug. A linear relationship was observed between the absorbance and the concentrations of the drug. The regression equation for the results was derived using the least square method. The plot (n = 3) was linear with very small intercept (0.0157) and good correlation coefficients (0.9984) in the concentration range of 5 - 40 µg/ml. The limit of detection (LOD) and limit quantitation (LOQ) were determined using the formula: LOD or LOQ = xSD/S , where x = 3 for LOD and 10 for LOQ. SD is the standard deviation of the intercept and S is the slope. The LOD and LOQ values were 0.24 µg/ml and 0.91 µg/ml respectively. The results are given in Table 1.

Table 1: Characteristics and statistical data for the regression equation of the proposed method

Parameter	Values
λmax (nm)	540
Beer's law limit (µg/ml)	5-40
Molar Absorptivity (L/mole/cm)	4.13 x 10^3
Colour Stability (h)	2
Regression equation (Y*)	
Slope (b)	0.9984
Intercept (a)	0.0157
Correlation coeffiecient (r)	0.9984
Limit of detection (µg/ml	0.24
Limit of quantitation (µg/ml)	0.91

**Y = a + bc, where c is the concentration of analyte (µg/ml) and Y is the absorbance unit.*

Precision: The precision of the proposed method was determined by replicate analysis of five separate samples of the standard solutions at a concentration level of 10 µg/ml. The relative standard deviation (RSD) was not more than 1.19%. This good precision level was suitable for quality control analysis of Rsperidone in its pharmaceutical dosage forms.

Interference studies: To explore the effect of some common excipients for the tablets on the analytical performance of the proposed method, samples were prepared by mixing known amount of Risperidone with various amounts of the excipients (Table 2). The analysis of these samples showed no interference was found from the tested excipients with the proposed method. The recovery values were 99.78±0.67 – 102.20±0.39 (Table 2). The obtained good recoveries ensured the suitability of the method for the analysis of dosage forms without interference from the common excipients.

Table 2: Analysis of Paroxetine in the presence of common excipients by the proposed method.

Excipients(*)	Recovery (%)±SD
Lactose (100)	102.20±0.39
Microcrystalline cellulose (10)	99.78±0.67
Magnesium sterate (10)	100.74±0.26
Sodium laury sulphate (10)	101.31±0.41
Starch (90)	99.85±0.84

Figures in parenthesis are the amounts in mg added per 20 mg of Paroxetine

Robustness and ruggedness: Robustness was determined by evaluating the effect of small variation in the experimental parameters on the analytical performance of the proposed method. In the study, one parameter was changed while others were kept constant and the recovery percentage was calculated each time. It was observed that none of these variables significantly affected the performance of the method. The recovery values 98.90±0.31 – 101.10±0.16 are shown in Table 3. This shows the reliability of the proposed method during routine application of the method. Ruggedness was assessed by applying the proposed method to the assay of Paroxetine using the same operational conditions on different days. Results obtained from day–to–day variations were reproducible as the RSD never exceeded 2%.

Table 3: Effect of small variation in the assay conditions on the analytical performance of the proposed spectrophotometric method for analysis of Paroxetine.

Parameter	Recovery (%)±SD
No variation	100.52±0.24
Chloranilic acid	
0.045	101.10±0.16
0.050	100.98±0.63
Reaction time (min)	
20	99.26±0.55
30	98.90±0.31

Assay of pharmaceutical bulk and dosage forms: It is evident from the results obtained previously that the proposed method as well as the official method gave satisfactory results with Paroxetine (bulk). The determination of Paroxetine in bulk by the proposed and BP method gave recoveries of 100.21±0.61 and 99.73±0.48 respectively. These results were compared with statistical analysis with respect to the accuracy (t - test) and precision (f - test) [32]. No significant differences were found between the calculated and theoretical values of *t*- and *F*-tests at 95% confidence level proving similar accuracy and precision in the determination of Paroxetine by both methods. This indicates the absence of any difference between the methods compared. However, as there was no official method for the analysis of the dosage forms, the tablets were subjected to the analysis of their contents of the studied drug only by the proposed method (Table 4). To establish further the suitability of the proposed method, samples were prepared by mixing known amounts of pure Paroxetine with some portions of powdered tablet dosage forms. The mixtures obtained were determined by the proposed method and the results are shown in Table 5.

Table 4: Determination of Paroxetine in pharmaceutical formulations (tablets) by the proposed method.

Volume of Risperidone solution/ml	Amount of Risperidone present/μg	Amount of Risperidone found/μg	Recovery (%)±SD
2	40	40.5818	101.45±0.93
3	60	59.8900	99.82±0.51
4	80	80.2768	100.35±0.82
5	100	100.1882	100.19±0.11

Values are mean of five determinations ± SD

Table 5: Recovery of Paroxetine in tablet – pure sample mixtures by the proposed method.

Risperidone In tablet (mg)	Pure risperdone added (mg)	Risperidone found (mg)	Recovery (%)±SD
20	0	20.005	100.25±0.60
20	2	22.121	100.55±0.98
20	4	24.098	100.41±0.73
20	6	25.963	99.86±0.81
20	8	28.134	100.48±0.49
20	10	29.891	99.64±0.51

Values are mean of three determinations ± SD

CONCLUSION

The proposed spectrophotometric method has the requisite accuracy, precision and sensitivity to assay Paroxetine in bulk and pharmaceutical solid dosage forms. The method lacks interferences from common excipients. Statistical evaluations of results in terms of t – and F–tests showed that the differences between the methods were insignificant and therefore the proposed method can be recommended for the routine analysis of Paroxetine in quality control laboratory.

REFERENCE

1. Mulliken RS. Structures of complex formed by halogens molecules with aromatic and oxygenated solvents. J. Amer. Chem. Soc. 1950. 72: 600 – 608.
2. Attama AA, Nnamai PO, Adikwu MU, Akidi FO. Spectrophotometric determination of haloperidol by charge transfer interaction with chloranilic acid. S. T. P. Pharma Science. 2008; 13(6): 419 – 421.
3. Karay AM, Wahbi AAM. Spectrophotometric assay of isoprenaline sulphate and methldopa using chloranil, analyst. 1979; 104, 146.
4. Taha MA, Ecrabat and Fatoda FA. Spectrophatometric assay of methylene tetramine, Analyst. 1980; 105, 568.
5. Abdee EE, Waley FM, Egindy AE, Wahbi AA. Spectrophotometric determination of flunarizine dihydrochloride through the formation of charge transfer complex with iodine. J. phab. Biomed. Analytical. 1995; 13, 58, 11.

6. Aespueiman and Tosvnsheade A. A spectrophotometric determination of microgram amounts of anion acids with chloranil, Analytical Chem., Acta. 1973; 66, 195.
7. Adikwu MU. Charge transfer complexation of chloranilic acid with pyrimethamine. Boll. Chem. Farmaceutico. 1996; 135, 128.
8. Agarwal SP, El-Sayed MA. Utility of acceptors in charge transfer complexation of alkaloids by chloranilic acid as a spectrophotometric titrant in non-aqueous media. Analyst, 1981; 106, 1157 – 1162.
9. Adikwu MU, Ofokansi KC. Spectrophotometric determination of moclobemide by charge transfer complexation. J. Pharm. Biomed. Anal. 1997; 16, 529 – 532.
10. Adikwu MU, Ofokansi KC, Attama AA. Spectrophotometric and thermodynamic studies of the charge transfer interaction between diethycarbamazine citrate and chloranilic acid acid. Chem. Pharm. Bull. 1999; 47(4), 463 – 466.
11. Adikwu MU, Ofokansi KC, Attama AA. Thermodynamic studies of the charge transfer interaction of chloranilic acid with moclobemide and promethazine hydrochloride. Biol. Pharm. Bull. 1998; 21(12): 1243 – 1246.
12. Barbui C, Furukawa TA, Cipriani A. Effectiveness of paroxetine in the treatment of acute major depression in adults;a systematic r-examination of published and unpublished data from randomized trials. Canadian Med. Assoc. J. 2008; 178(3): 296 – 305.
13. Baldwin DS, Anderson IM, Nutt DJ, Bandelowa B, Bond A, Davidson JR, dan Boer JA, Fineberg N A, Knapp M, Scott J. Wittchen HU. Evidence-based guidelines for the pharmacological treatment of anxiety disorder: recommendations from the British Association of Psychopharmacology. 2005; 19(6): 567 – 596.
14. Nouws HPA, Delerue-Matos C, Barros AA, Rodrigues JA. Electroanalytical determination of paroxetine in pharmaceuticals. J. Pharma. and Biomed. Anal. 2006; 42(3): 341–346,
15. Erk N. Biryol J. Voltammetric and HPLC techniques for the determination of paroxetine hydrochloride. Pharmazie. 2003; 58(10): 699–704.
16. Robert S, Genowefa M, Marcin K. Determination of fluoxetine and paroxetine in pharmaceutical formulations by densitometric and videodensitometric TLC. J. Planar Chromatogr. Modern TLC, 2003; 16(1): 19–22.
17. Venkatachalam A, Chatterjee VS. Stability-indicating high performance thin layer chromatography determination of Paroxetine hydrochloride in bulk drug and pharmaceutical formulations. Analytica Chimica Acta. 2007; 598(2): 312–317.

18. Zainaghi IA, Lanchote VL, Queiroz RHC. Determination of paroxetine in geriatric depression by high-performance liquid chromatography. Pharmacological Research. 2003; 48(2): 217–221.
19. Zhu Z, Neirinck L. High-performance liquid chromatography-mass spectrometry method for the determination of paroxetine in human plasma. J. Chromatogr. B. 2002; 780(2): 295–300.
20. Massaroti P, Cassiano NM, Duarte LF, et al. Validation of a selective method for determination of paroxetine in human plasma by LC-MS/MS. J. Pharm. and Pharmaceu. Sci. 2005; 8(2) 340–347.
21. Jhee OH, Seo HK, Lee MH, et al. Determination of paroxetine in plasma by liquid chromatography coupled to tandem mass spectrometry for pharmacokinetic and bioequivalence studies. Arzneimittel-Forschung. 2007; 57(7): 455–461.
22. British Pharmacopoeia. The Stationary Office, London, UK. 2003.
23. United States Pharmacopeial Convention. The United States Pharmacopeia 31, The National Formulary 26, United States Pharmacopeia, Rockville, Md, USA. 2008.
24. Eap CB, Bouchoux G, Amey M, Cochard N, Savary L, Baumann P. Simultaneous determination of human plasma levels of citalopram, paroxetine, sertraline, and their metabolites by gas chromatography-mass spectrometry. J. Chromatogr. Sci. 1998; 36(7): 365–371.
25. Hans JL, Werner W, Günter F. Improved sample preparation for the quantitative analysis of paroxetine in human plasma by stable isotope dilution negative ion chemical ionisation gas chromatography-mass spectrometry. J. Chromatogr. B. 2002; 779(2): 353–357.
26. Chien L, Emily SG, Sidney HK, Alan N, Ronald TC, Glen BB. Determination of paroxetine levels in human plasma using gas chromatography with electron-capture detection. J. Chromatogr. B. 2000; 749(2): 275–279.
27. Labat L, Deveaux M, Dallet P, Dubost JP. Separation of new antidepressants and their metabolites by micellar electrokinetic capillary chromatography. J. Chromatogr. B. 2002; 773(1): 17–23.
28. Onal A, Kepekci SE, Oztunc A. Spectrophotometric methods for the determination of the antidepressant drug paroxetine hydrochloride in tablets. J. AOAC Int’l. 2005; 88(2): 490 – 495.
29. Darwish IA, Refaat IH. Spectrophotometric analysis of selective serotonin reuptake inhibitors based on formation of charge transfer complexes with tetracyanoquinodimethane and chloranilic acid. J. AOAC Int’l. 2006; 89(2): 326 - 333

30. Darwish IH. Development and validation of spectrophotometric methods for determination of fluoxetine, sertraline and paroxetine in pharmaceutical osage forms. J. AOAC Int'l. 2005; 88(1): 38 – 45.
31. Rose J. Advanced Physicochemical Experiments, Pitman, London, 1964; 67.
32. Rouessac F, Rouessac A. Chemical Analysis Modern Instrumentation Methods and Techniques, 4th ed. John Wiley and Sons, New Jersey. 2004; 390.