Spectrofluorimetric Determination of Citalopram and Desvenlafaxine Antidepressant Drugs in Micellar Medium

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Abstract

A spectrofluorimetric method for the determination of antidepressant drugs namely citalopram (CIT) and desvenlafaxine (DSV) in tablets has been developed. The method is based on native fluorescence of these drugs in micellar solution of sodium dodecyl sulfate (SDS). The wavelengths of excitation and emission were 240 nm and 595 nm for citalopram and 225 nm and 600 nm for desvenlafaxine. The method allows the determination of 0.8-8.0 μg mL^{-1} of citalopram and 0.4-4.0 μg mL^{-1} of desvenlafaxine in presence of 18 mM SDS, in an acetate-HCl buffer solution of pH=5. The limits of detection (LOD) were 0.043 and 0.034 μg mL^{-1} for CIT and DSV, respectively. The fluorescence properties of CIT and DSV in micellar media were also studied. This method has been successfully applied to determination of the two drugs in tablets with satisfactory results.

1. Introduction

Citalopram, (CIT, Fig. 1a) 1-(3-dimethylaminopropyl)-1-(4-fluorophenyl)-1, 3-dihydroisobezofuran-5-carbonitrile [1], is a second generation antidepressant and one of the recently introduced SSRIs. It is used for managing depression, social anxiety disorder, panic disorder and obsessive-compulsive disorder [2-4]. Several methods have been devised for the determination of citalopram in pharmaceutical preparations and biological fluids. These include high performance liquid chromatography (HPLC) with UV detectors [5-7], HPLC with fluorescence detectors [8-10] HLPC/mass spectrometry [11-13], gas chromatography [14,15], electrophoretic methods [16-18] and spectrophotometric methods [19-21]. However, few spectrofluorimetric methods [22-24] have been reported in the literature, and most reported methods involve multistep procedures, and have poor selectivity's and sensitivities. Desvenlafaxine succinate is a newer antidepressant drug, which is chemically 1-[(1RS)-2- (Dimethylamino)-1-(4-hydroxyphenyl) ethyl] cyclohexanol succinate monohydrate (DSV, Fig. 1b). Desvenlafaxine succinate is a structurally novel SNRI (serotonin - norepinephrine reuptake inhibitor) useful for the treatment of MDD (major depressive disorder). Desvenlafaxine (O-desmethyl venlafaxine) is the major active metabolite of the antidepressant venlafaxine, a medication used to treat major depressive, generalized anxiety and panic disorders. Literature survey reveals HPLC coupled to spectrophotometric [25-28], spectrofluorimetric [29-32] or coulometric detection [33], capillary electrophoresis [34-38], adsorptive stripping voltammetric [39], HPLC-ESI/MS [40,41] and LC-MS/MS [42-46], have been used. Desvenlafaxine succinate is not official in any pharmacopoeia. In the present paper, the fluorescence characteristics of citalopram and desvenlafaxine succinate are investigated in micellar media. Based on obtained results, sensitive micelle-enhanced spectrofluorimetric method has been firstly developed for the determination of citalopram and desvenlafaxine succinate drugs in bulk and tablet samples.
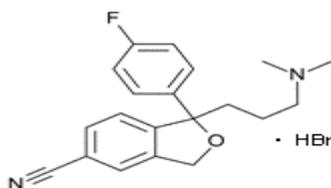
a- Citalopram
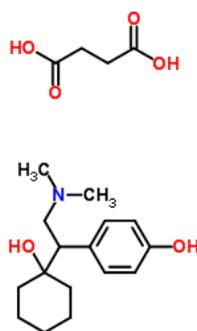
b- Desvenlafaxine succinate

Fig. 1. Chemical structure of the studied drugs.

2. Materials and Methods

2.1. Apparatus

Spectrofluorimetric measurements were carried out on a Shimadzu RF-5301 PC spectrofluorimeter (Kyoto, Japan), equipped with a 150 W xenon lamp and using 1.0 cm quartz cells. The excitation and emission monochromators were fixed with 1.5 mm slits. The pH values of buffer solutions were measured using Jenway instrument pH-meter (combined electrode).

2.2. Reagents and Materials

i. Preparation of standard solution: Pharmaceutical grade of CIT and DSV certified to be 99.85% pure was obtained as gift were kindly supplied from Egyptian International Pharmaceutical Industries Company (EIPICo), Egypt. Stock solutions of pure CIT and DSV were prepared separately by dissolving 10 mg (accurately weighed) of each drug in least amount of water and finally the volume was made up to 100 mL with distilled water (100 $\mu g\ mL^{-1}$).

ii. Series of buffer solutions of KCl-HCl (pH=1.0–2.2), potassium hydrogen phthalate-HCl (pH =2.2–4.0) and NaOAc-HCl (pH = 3.2–6.8) were prepared by standard methods.

iii. Solutions of 0.1M sodium dodecyl sulphate (SDS), sodium dodecyl benzene sulphonate (SDBS), cetyl trimethylammonium bromide (CTAB), cetyl pyridinium bromide (CPB), tween 80 and triton-X (TX-100) were separately prepared.

All reagents used were of analytical reagent grade purity or of high grade purity.

2.3. General Procedure and Calibration Graphs

Into a set of 25 mL calibrated flasks, an aliquot of working standard solution of each drug was added and then following solutions were added in sequence: 2 mL of acetate buffer solution of pH=5 and 2 mL of 0.1M SDS solution. The resultant mixture was diluted to the mark with distilled water, mixed thoroughly by shaking and then allowed to stand for 10 minutes. A blank sample containing all the reagents except the drug was also prepared. The fluorescence intensities of the sample and blank were then measured in a 10 mm path-length quartz cell.

2.4. Procedure for Tablets

At least ten tablets of the drugs were weight into a small dish, powdered and mixed well. A portion equivalent to 10 mg of CIT and DSV were weight and dissolved in distilled water, filtered into a 100 mL calibrated flask and diluted to volume with water. Solutions of working range concentration were prepared by proper dilution of this stock solution with water and followed the above procedure for calibration curves. The nominal content of the tablets was calculated either from a previously plotted calibration graphs or using the regression equation.

3. Results and Discussion

3.1. Fluorescence Spectra

Both citalopram and desvenlafaxine show native fluorescence in an aqueous solution. The fluorescence spectra of both the drugs in aqueous solution and micellar medium were recorded (Figs. 2, 3). For citalopram, an excitation wavelength was 240 nm and an emission maximum was obtained at 595 nm (Fig. 2). For desvenlafaxine, an excitation wavelength was 225 nm and an emission maximum was obtained at 600 nm (Fig. 3).
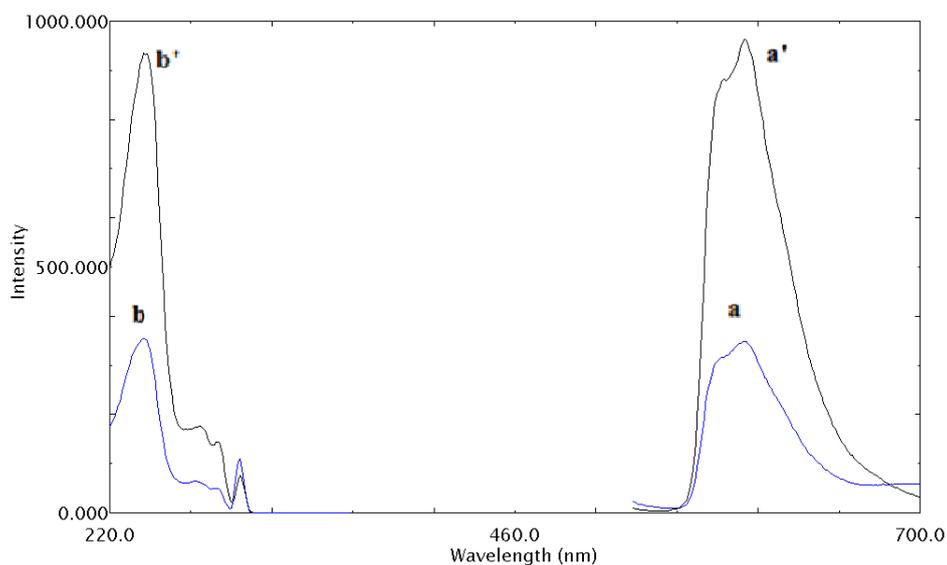
Fig. 2. Emission spectra (a, ȧ) and excitation spectra (b, ḃ) of (8.0 µg mL^{-1}) CIT: (ȧ, ḃ) in micellar medium of SDS and (a, b) in aqueous solution at pH = 5.

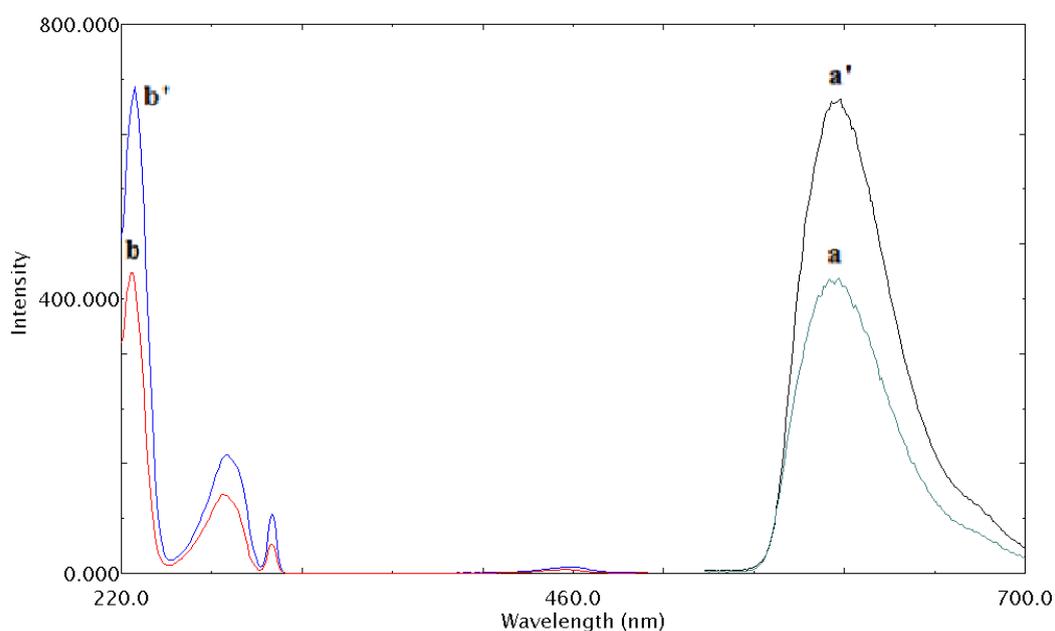**Fig. 3**. Emission spectra (a, ȧ) and excitation spectra (b, ḃ) of (4.0 µg mL^{-1}) DVS: (ȧ, ḃ) in micellar medium of SDS and (a, b) in aqueous solution at pH = 5.

3.2. Effects of pH and Buffers

The effect of pH on the enhanced fluorescence of drugs in micellar medium was studied. The results showed the optimum pH=5 for both citalopram and desvenlafaxine (Fig. 4). Different buffer solutions (acetate, phosphate and Britton Robinson buffer) were tested. NaOAc-HCl buffer solution was found to be most successful for both drugs. So NaOAc–HCl buffer (pH=5.0) is selected for the assay and the optimum volume of NaOAc–HCl buffer is 2 mL.
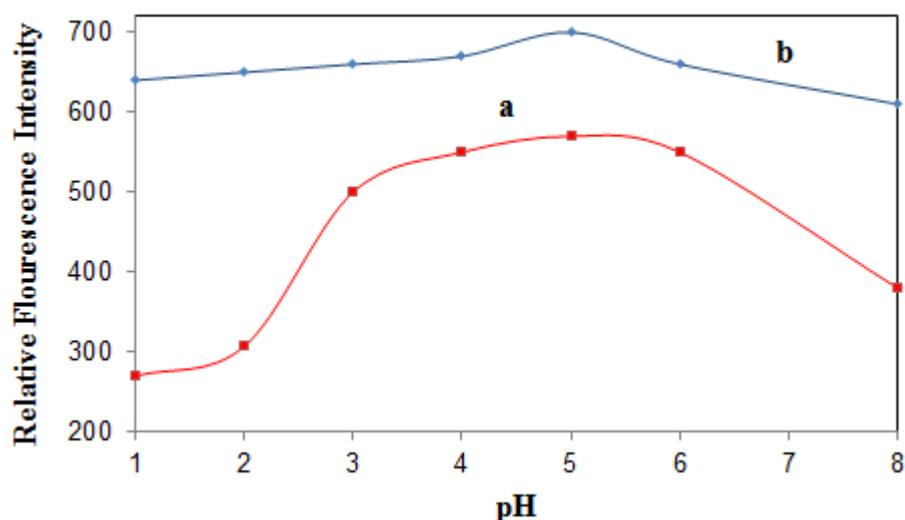
Fig. 4. Effect of pH values on the fluorescence intensity of a: (4 µg mL^{-1}) CIT and b: (4 µg mL^{-1}) DVS in SDS.

3.3. Effect of surfactants

Addition of a surfactant above its critical micelle concentration (CMC) increases the fluorescence intensity of many fluorophores. This fact has been used to develop improved methods for spectrofluorimetric determination of many compounds [47,48]. In the present paper, the effect of six surfactants; an anionic sodium dodecyl sulphate (SDS), sodium dodecyl benzene sulphonate (SDBS), a cationic cetyl trimethylammonium bromide (CTAB), cetyl pyridinium bromide (CPB), and a neutral surfactant tween 80 and TritonX–100 (TX–100) on fluorescence intensity of citalopram and desvenlafaxine were investigated. Figure 5 shows that the enhancement of the fluorescence intensity of both drugs was maxima in SDS solution and therefore SDS was chosen for further study.
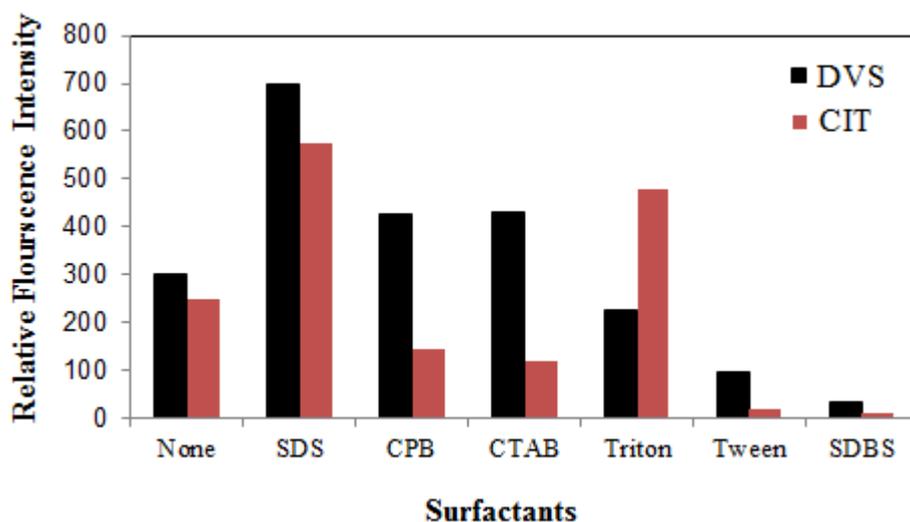
Fig. 5. Effect of different surfactants on fluorescence intensity.

3.4. Effect of SDS Concentration

The effect of SDS concentration on fluorescence of both the drugs was studied. With increase in SDS concentration, the fluorescence intensity increased and reached a stable value at 14 mM concentration of SDS which is above the CMC value of 6 mM for SDS (Fig. 6). Further increase in concentration of SDS produced little variation in fluorescence

intensity. Therefore, 18 mM concentration was selected as the optimum concentration of SDS for carrying out further investigations.
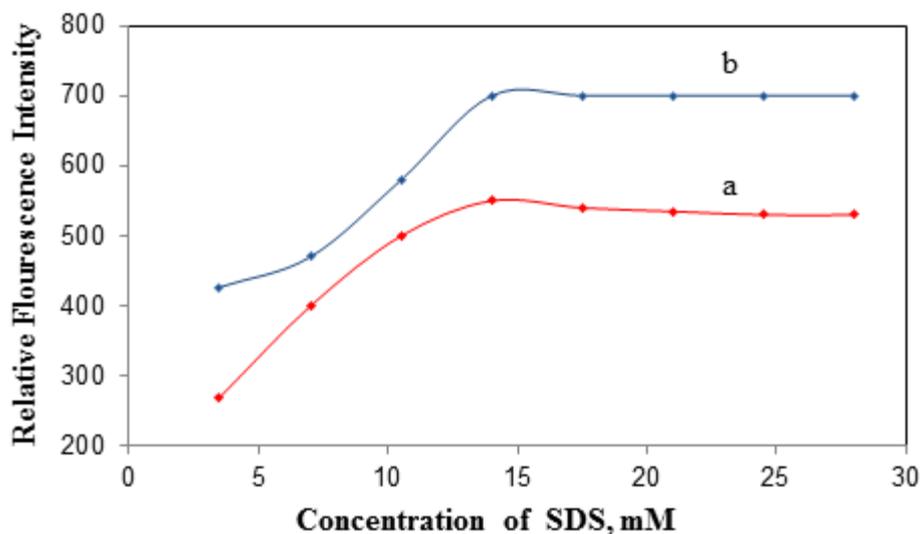

Fig. 6. Effect of SDS concentration on the fluorescence intensity of a: (4 µg mL^{-1}) CIT and b: (4 µg mL^{-1}) DVS, at pH=5.

3.5. Effect of Diluting Solvents

Effect of diluting solvent was also tested using different solvents such as, water, ethanol, methanol, acetonitrile, butanol and DMF (Fig. 7). Using water as diluting solvent gives the highest fluorescence intensity value. Other solvents as butanol and DMF showed a distinct sharp decrease in the fluorescence intensity.
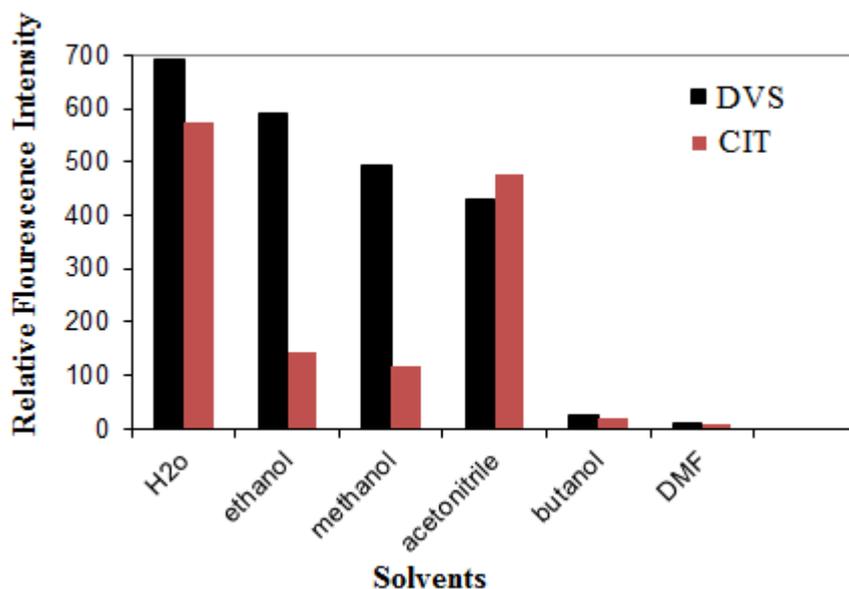

Fig. 7. Effect of different solvents on fluorescence intensity.

3.6. Sequence of Addition

A series of solutions were prepared by changing the sequence of addition of reagents while keeping the same concentration and fluorescence intensity was measured. It was observed that change in sequence of addition of the

reagents produced little variation in fluorescence intensity. The optimal order of addition of reagents selected was: drug, buffer and SDS.

3.7. Effect of Temperature

The effect of temperature on the fluorescence intensity of the CIT and DSV in micellar medium was carried out at different temperature settings (room temperature, 40, 60, 80, and 100°C) using a thermostatically controlled water bath. Maximum and constant fluorescence intensity was obtained at room temperature. This effect can be explained by higher internal conversion as temperature increases, facilitating non-radiative deactivation of excited singlet state [49]. A temperature of 25 °C, close to room temperature, was selected for carrying out further experiments. The induced fluorescence intensity was found stable for more than 3 h.

3.8. Luminescence Mechanism

Citalopram and desvenlafaxine are hydrophobic drugs having low polarity. In the presence of micellar medium of SDS the solubility of the drug in the micellar medium is greatly increased. Due to change in microenvironment of the drug, there is decrease in non-radiative energy loss through molecular collisions and quantum efficiency of fluorescence is greatly increased [50]. The optimum concentration of SDS is above the CMC value (18 mM) which indicates that formation of micelles greatly increases the fluorescence intensity of the system.

4. Validation of the Method

Validation of optimized proposed method was done with respect to following parameters.

4.1. Linearity and Range

Under the described experimental conditions, the fluorescence intensity-concentration plot was rectilinear over the range of 0.8-8.0 µg mL^{-1} for CIT and over the range of 0.4-4.0 µg mL^{-1} for DSV (Table 1). The dependence of the relative fluorescence intensity on the concentration of CIT and DSV shown in Fig. 8. The relative fluorescence intensity increased with the increase of drugs concentration.
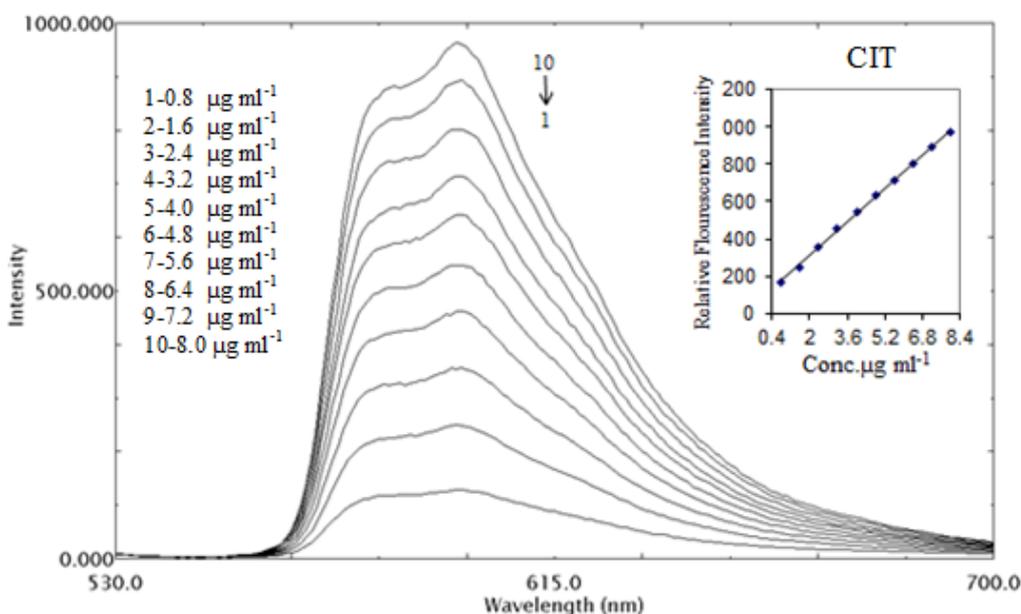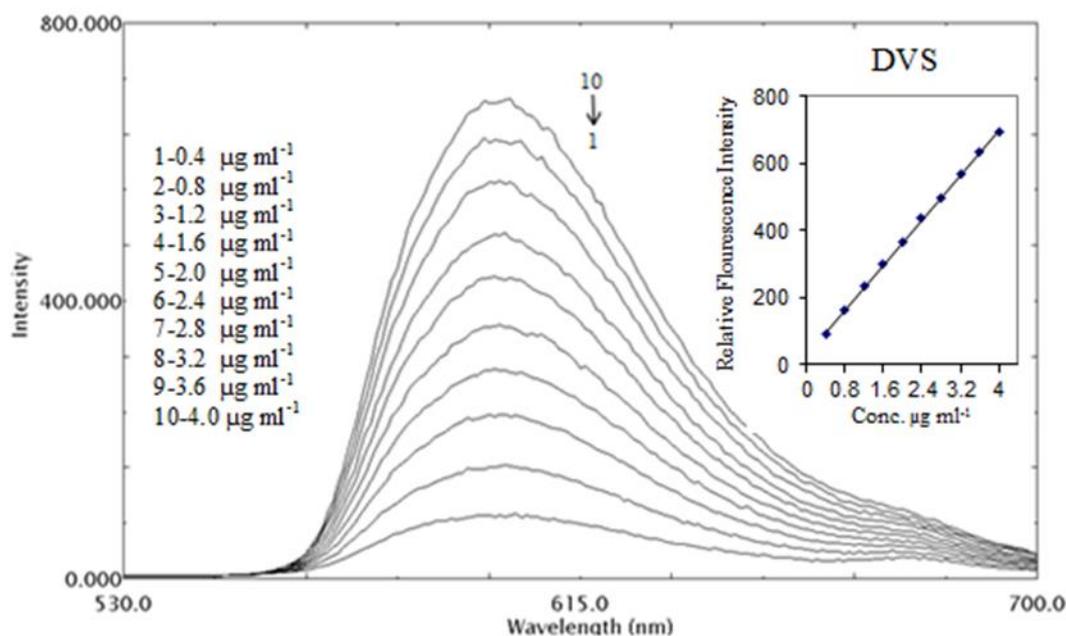
Fig. 8. Emission spectra in micellar system with different concentration of CIT and DSV at pH=5.

Table 1. Statistical and analytical parameters of the proposed method.

Parameters	Drugs	
	CIT	DSV
λ_{em} (nm)	595	600
λ_{ex} (nm)	240	225
Beer's law limit (μg mL^{-1})	0.8-8.0	0.4-4.0
Correlation coefficient (r)	1.000	0.9997
Linear regression equation		
$\mathbf{S_{y/x}}$	5.933	3.273
Intercept (a)	60.933	27.446
Slope (b)	120.857	170.121
S.D of slope (S_b)	1.772	1.956
S.D. of intercept (S_a)	5.523	3.047
Detection Limits (μg mL^{-1})	0.0439	0.0344
Quantitation Limits (μg mL^{-1})	0.1466	0.1149

F = a + bC, where F is the fluorescence intensity and C is the concentration of drug in μg mL^{-1}

4.2. Limit of Detection and Limit of Quantitation

The detection and quantification limits as defined by IUPAC [51], LOD $=3S_b/m$ and LOQ $=10S_b/m$ (where S_b is the standard deviation of the blank and m is the slope of the calibration graph) were found to be 0.0439 and 0.0344 μg mL^{-1}, 0.1466 and 0.1149 μg mL^{-1} for CIT and DSV, respectively (Table 1). The slope of the calibration graph (m) is the calibration sensitivity according to IUPAC definition.

4.3. Accuracy and Precision

In order to determine the accuracy and precision of the recommended procedure five replicate determinations at three different concentrations of the studied drugs were carried out. Precision and accuracy were based on the calculated

relative standard deviation (RSD, %) and relative error (RE, %) of the found concentration compared to the theoretical one, respectively (Table 2) and indicate that the proposed method is highly accurate and reproducible.

Table 2. Evaluation of accuracy and precision of the proposed method.

Drugs	Drug Taken µg mL^{-1}	Drug Found µg mL^{-1}	Recovery[c], %	RE[a]	RSD[b], %	SE
CIT	1.6	1.5993	99.96	-0.044	1.663	2.049
	4.0	3.9992	99.98	-0.021	0.826	2.274
	6.4	6.3961	99.94	-0.061	0.510	2.049
DVS	0.8	0.7995	99.94	-0.063	4.443	3.563
	2.0	1.9998	99.99	-0.010	1.744	3.134
	3.2	3.1987	99.96	-0.041	1.349	3.791

[a]Relative error. [b]Relative standard deviation. [c]Mean value of five determinations.

4.4. Selectivity

The selectivity of the proposed method was checked by investigating the interference effect of common excipients such as talc, sucrose, lactose, starch and magnesium stearate on the analysis of CIT and DSV. No interferences were observed in the determination of the investigated drugs in the presence of the common excipients (Table 3) and average recoveries obtained were found in the range of 99.94 to 99.99%.

Table 3. Determination in CIT and DSV in pharmaceutical preparations.

Drugs	Drug Taken µg mL^{-1}	Drug Found µg mL^{-1}	Recovery[c], %	RE[a]	RSD[b], %
CIT (Cipramax[d] 40 mg)	1.6	1.599	99.96	-0.044	1.881
	4.0	3.998	99.96	-0.040	1.171
	6.4	6.397	99.96	-0.041	0.824
DVS (Prismaven[e] 100 mg)	0.8	0.799	99.99	-0.013	4.797
	2.0	1.998	99.94	-0.060	1.216
	3.2	3.198	99.94	-0.063	1.433

[a]Relative error. [b]Relative standard deviation. [c]Mean value of five determinations.

[d]Cipramax tablets (copad egypt for trade and pharmaceutical industries, Egypt).

[e]Prismaven (rameda for pharmaceutical industries, 6th of october city, Egypt).

4.5. Analysis of Marketed Formulations

The proposed method was applied to the determination of the studied drugs in conventional tablets. The results obtained are satisfactorily accurate and precise as indicated by the excellent % recovery as shown in (Table 3). The statistical analysis of the results detected no significant difference in the performance of the proposed methods and the reported methods by the Student's t-value and variance ratio F-value. The results of this study are given in Table 4.

Table 4. Analysis of tablets containing CIT and DVS using the proposed method and reported methods

Drug	Drug formulation	Labeled amount	%Recovery±SD[a]		t-test[b]	F-test[b]	Ref.
			Proposed	Reported			
CIT	Cipramax	40 mg/tab	99.96±0.092	100.03±0.101	1.701	1.205	20
DVS	Prismaven	100 mg/tab	99.96±0.887	99.56±1.095	2.520	1.523	41

[a]Average of five determinations.
[b]Tabulated values at 95% confidence limit are t=2.306, F=6.338.

5. Conclusion

A simple and non-extractive spectrofluorimetric method was developed for the determination of CIT and DSV in a micellar medium. The method is fast, sensitive and has low detection limit and can be applied to the determination of the studied drugs in dosage forms.

References

[1] S. Budavari, The Merck Index, 14th edn. Merck and Co., Inc, Whitehouse Station, (2007), 387.

[2] L.L. Brunton, K.L. Parker, D.K. Blumenthal, I. Buxton, Goodman & Gilman's. The pharmacological basis of therapeutics. McGraw-Hill, New York, (2008).

[3] In: http://en.wikipedia.org/wiki/Citalopram.

[4] S.C. Sweetman, Martindale: The complete drug reference. Pharmaceutical Press, London, (2005).

[5] O.V. Olesen, K. Linnet, Simplified high-performance liquid chromatographic method for the determination of citalopram and desmethylcitalopram in serum without interference from commonly used psychotropic drugs and their metabolites, J. Chromatogr. B 675 (1996) 83–88.

[6] C. Duverneuil, G.L. De La Grandmaison, P. De Mazancourt, J.C. Alvarez, A high performance liquid chromatography method with photodiode-array UV detection for therapeutic drug monitoring of the nontricyclic antidepressant drugs, Ther. Drug Monit. 25 (2003) 565–573.

[7] C. Frahnert, M.L. Rao, K. Grasmader, Analysis of eighteen antidepressants, four atypical antipsychotics and active metabolites in serum by liquid chromatography: a simple tool for therapeutic drug monitoring, J. Chromatogr. B 794 (2003) 35–47.

[8] E. Matsui, M. Hoshino, A. Matsui, A. Okahira, Simultaneous determination of citalopram and its metabolites by high performance liquid chromatography with column switching and fluorescence detection by direct plasma injection, J. Chromatogr. B: Biomed. Appl. 668 (1995) 299–207.

[9] B. Rochat, M. Amey, P. Baumann, Analysis of enantiomers of citalopram and its demethylated metabolites in plasma of depressive patients by chiral reverse-phase liquid chromatography, Ther. Drug Monit. 17 (1995) 273–279.

[10] Q. Meng, D. Gauthier, Simultaneous analysis of citalopram and desmethylcitalopram by liquid chromatography with fluorescence detection after solid-phase extraction, Clin. Biochem. 38 (2005) 282–285.

[11] H. Juan, Z. Zhiling, L. Huande, Simultaneous determination of fluoxetine, citalopram, paroxetine, venlafazine in plasma by high performance liquid chromatography-electrospray ionization mass spectrometry (HPLCC-MS/ESI),   J. Chromatogr. B 820 (2005) 33–39.

[12] U. Gutteck, K.M. Rensch, Therapeutic drug monitoring of 13 antidepressant and five neuroleptic drugs in serum with liquid chromatography-electrospray ionization mass spectrometry, Clin. Chem. Lab. Med. 41 (2003) 1571–1579.

[13] M. Kollroser, C. Schober, An on-line solid phase extraction liquid chromatography-tandem mass spectrometry method for the analysis of citalopram, fluvoxamine and paroxetine in human plasma, Chromatographia 57 (2003) 133–138.

[14] M. Gergov, J. Nokua, I. Rasanen, I. Ojenpera, Validation and quality assurance of a broad scale gas-chromatographic screening method for drugs, Z. Zagadnien Nauk. Sadowych. 43 (2000) 70–78.

[15] R.T. Sane, M. Francis, B.S. Dawkhar, Quantitative gas chromatographic determination of citalopram in pharmaceutical formulations, Indian Drugs 39 (2002) 525–528.

[16] J.R. Flores, J.J.B. Nevado, A.M.C. Salcedo, M.P.C. Diaz, Development of capillary zone electrophoretic method to determine six antidepressants in their pharmaceutical preparations experimental design for evaluating the ruggedness of method, J. Sep. Sci. 27 (2004) 33–40.

[17] T. Buzinkaiova, J. Polonsky, Determination of four selective serotonin reuptake inhibitors by capillary isotachophoresis, Electrophoresis 21 (2000) 2839–2841.

[18] A. Bjorhovde, T.G. Halvorsen, K.E. Rasmussen, S. Pedersen-Bjergaard, Liquid-phase microextraction of drugs from human breast milk, Anal. Chim. Acta 491 (2003) 155–161.

[19] R. Asad, Development and application of spectrophotometric methods for the determination of citalopram hydrobromide in dosage forms, Chem. Pharm. Bull. 54 (2006) 432–434.

[20] B. Tirupathi and G. Venkateshwarlu, Oxidative spectrophotometric determination of drugs by using cerium (IV) and rhodamine-B dye. Asian J. Biochem. Pharm. Res. 2015, 5, 1-11

[21] N. Badiadka, V. Kunnummel, Spectrophotometric determination of citalopram hydrobromide in pharmaceuticals, J. Mex. Chem. Soc. 54 (2010) 98–102.

[22] S.G. Vasantharaju, S.L. Prabu, A. Jacob, Spectrofluorimetric method for determination of citalopram in bulk and pharmaceutical dosage forms, Indian J. Pharm. Sci. 70 (2008) 647–648.

[23] H.E. Satana, E. Nusret, G.G. Nilgun, Spectrofluorimetric determination of citalopram HBr in tablets, FABAD J. Pharm. Sci. 32 (2007) 73–77.

[24] D.T. El-Sherbiny, Spectrofluorimetric determination of citalopram in pharmaceutical preparations and spiked human plasma using organized media,   J. AOAC. Int. 89 (2006) 1288–1295.

[25] S.M. Pawar, L.D. Khatal, S.Y. Gabhe, S.R. Dhaneshwar, LC–UV and LC–MS evaluation of stress degradation behavior of desvenlafaxine, J. Pharm. Anal. 2 (2012) 264–271.

[26] M. Matoga, F. Pehourcq, K. Titier, F. Dumora, C. Jarry, Rapid high-performance liquid chromatographic measurement of venlafaxine and *O*-desmethylvenlafaxine in human plasma: Application to management of acute intoxications, J. Chromatogr. B: Biomed. Sci. Appl. 760 (2001) 213–218.

[27] S.M. Pawar, S.R. Dhaneshwar, Application of stability indicating high performance thin layer chromatographic method for quantitation of desvenlafaxine in pharmaceutical dosage form, J. Liq. Chromatogr. Related Technol. 35 (2012) 499–510.

[28] D.R. Hicks, D. Wolaniuk, A. Russell, N. Cavanaugh, M. Kraml, A high-performance liquid chromatographic method for the simultaneous determination of venlafaxine and o-desmethylvenlafaxine in biological fluids, Ther. Drug Monit. 16 (1994) 100–107.

[29] K.H. Ardakani, A. Foroumadi, M.R. Rouini, Development and validation of a rapid HPLC- fluorescence method for simultaneous determination of venlafaxine and its major metabolites in human plasma, Daru 18 (2010) 97–102.

[30] M. Roberto, M. Laura, C. Roberta, F. Salvatore, A. Mario, A.R. Maria, Analysis of the second generation antidepressant venlafaxine and its main active metabolite *O*-desmethylvenlafaxine in human plasma by HPLC with spectrofluorimetric detection, J. Chromatogr. B 856 (2007) 88–94.

[31] R. Waschgler, W. Moll, P. König, A. Conca, Therapeutic drug monitoring of trazodone: are there pharmacokinetic interactions involving citalopram and fluoxetine, Int. J. Clin. Pharmacol. Ther. 42 (2004) 120–124.

[32] R.L. Vu, D. Helmeste, L. Albers, C. Reist, Rapid determination of venlafaxine and O-desmethylvenlafaxine in human plasma by high-performance liquid chromatography with fluorimetric detection, J. Chromatogr. B: Biomed. Sci. Appl. 703 (1997) 195–201.

[33] S. Fanali, P. Catarcini, M.G. Quaglia, Use of vancomycin silica stationary phase in packed capillary electrochromatography: III. Enantiomeric separation of basic compounds with the polar organic mobile phase, Electrophoresis 23 (2002) 477–485.

[34] L. Labat, M. Deveaux, P. Dallet, J.P. Dubost, Separation of new antidepressants and their metabolites by micellar electro kinetic capillary chromatography, J. Chromatogr. B: Analyt. Technol. Biomed. Life Sci. 773 (2002) 17–23.

[35] C. Desiderio, Z. Aturki, S. Fanali, Use of vancomycin silica stationary phase in packed capillary electrochromatography I. Enantiomer separation of basic compounds, Electrophoresis 22 (2001) 535–543.

[36] S. Fanali, S. Rudaz, J.L. Veuthey, C. Desiderio, Use of vancomycin silica stationary phase in packed capillary electrochromatography: II. Enantiomer separation of venlafaxine and *O*-desmethylvenlafaxine in human plasma, J. Chromatogr. A 919 (2001) 195–203.

[37] S. Rudaz, C. Stella, E. Balant-Gorgia, S. Fanali, J.L. Veuthey, Simultaneous stereoselective analysis of venlafaxine and *O*-desmethylvenlafaxine enantiomers in clinical samples by capillary electrophoresis using charged cyclodextrins, J. Pharm. Biomed. Anal. 23 (2000) 107–115.

[38] S. Rudaz, J.L. Veuthey, C. Desiderio, S. Fanali, Enantioseparation of venlafaxine and*O*-desmethylvenlafaxine by capillary electrophoresis with mixed cyclodextrins, Chromatographia 50 (1999) 369–372.

[39] B.J. Sanghavi, A.K. Srivastava, Adsorptive stripping differential pulse voltammetric determination of venlafaxine and desvenlafaxine employing nafion–carbon nanotube composite glassy carbon electrode, Electrochim. Acta 56 (2011) 4188–4196.

[40] W. Liu, H.L. Cai, H.D. Li, High performance liquid chromatography-electrospry ionization mass spectrometry (HPLC-MS/ESI) method for the simultaneous determination of venlafaxine and its three metabolities in human plasma, J. Chromatogr. B: Analyt. Technol. Biomed. Life Sci. 850 (2007) 405–411.

[41] Bahia Abbas Moussa, Ramzia Ismail El-Bagary and Yasmin Abdullah Al-Eryani Development and Validation of a Stability-Indicating HPLC Method for the Analysis of Desvenlafaxine Succinate in the Presence of its Acidic Induced Degradation Product in Bulk and Pharmaceutical Preparation. *J. Chem. Pharm. Res.,* **2011**, 3, p. 425-437

[42] G.R. Shah, B.T. Thaker, K.R. Surati, M.H. Parabia, Simultaneous determination of venlafaxine and its main active metabolite *o*-desmethyl venlafaxine in rat plasma by LC-MS/MS, Anal. Sci. 25 (2009) 1207–1210.

[43] D. Rajasekhar, I.J. KumarJ, P. Venkateswarlu, Rapid high-performance liquid chromatography–tandem mass spectrometry method for simultaneous measurement of venlafaxine and *O*-desmethylvenlafaxine in human plasma and its application in comparative bioavailability study, Biomed. Chromatogr. 23 (2009) 1300–1307.

[44] A. de Castro, M. Concheiro, O. Quintela, A. Cruz, M. LÓpez-Rivadulla, LC–MS/MS method for the determination of nine antidepressants and some of their main metabolites in oral fluid and plasma: Study of correlation between venlafaxine concentrations in both matrices, J. Pharm. Biomed. Anal. 48 (2008) 183–193.

[45] B.N. Patel, N. Sharma, M. Sanyal, P.S. Shrivastav, Liquid chromatography tandem mass spectrometry assay for the simultaneous determination of venlafaxine and *O*-desmethylvenlafaxine in human plasma and its application to a bioequivalence study, J. Pharm. Biomed. Anal. 47 (2008) 603–611.

[46] N. Castaing, K. Titier, M. Receveur-Daurel, M. Le-Deodic, D. Le-Bars, N. Moore, M. Molimard, Quantification of eight new antidepressants and five of their active metabolites in whole blood by high-performance liquid chromatography-tandem mass spectrometry, J. Anal. Toxicol. 31 (2007) 334–341.

[47] J.A. Ocana, F.J. Barragan, M. Callejon, Fluorescence and terbium-sensitised luminescence determination of garenoxacin in human urine and serum, Talanta 63 (2004) 691–697.

[48] A.M. Garcia-Campana, J.J. Aaron, J.M. Bosque-Sendra, Micellar-enhanced photochemically induced fluorescence detection of chlorophenoxyacid herbicides: flow injection analysis of mecoprop and 2,4-dichlorophenoxyacetic acid, Talanta 55 (2001) 531–539.

[49] W.R. Seitz, in: P.J. Elving, E.J. Meehan, I.M. Kolthoff (Eds), Treatise on analytical chemistry, vol 7 (Ch 4), Wiley, New York, (1981).

[50] C.L. Tong, Y. Zhu, W.P. Liu, Study on the co-luminescence system of Dy-Gd-1,6-bis (1-phenyl-3-methyl-5-pyrazol-4-one) hexanedione-cetyltrimethyl ammonium bromide and its analytical application, Analyst 126 (2001) 1168–1171.

[51] J.D. Winefordner, G.L. Long, Limit of detection: a closer look at IUPAC definition, Anal. Chem. 55 (1983) 712A–724A.

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