DRUGS, COSMETICS, FORENSIC SCIENCES

Spectrophotometric Analysis of Selective Serotonin Reuptake Inhibitors Based on Formation of Charge-Transfer Complexes with Tetracyanoquinodimethane and Chloranilic Acid

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A simple, accurate, and sensitive spectrophotometric method for analysis of selective serotonin reuptake inhibitors (SSRIs) has been developed and validated. The analysis was based on the formation of colored charge-transfer complexes between the intact molecule of SSRI drug as an *n*-electron donor and each of tetracyanoquinodimethane (TCNQ) or *p*-chloranilic acid (pCA) as electron acceptors. The formed complexes were measured spectrophotometrically at 842 and 520 nm for TCNQ and pCA, respectively. Different variables and parameters affecting the reactions were studied and optimized. Under the optimum reaction conditions, linear relationships with good correlation coefficients (0.9975–0.9996) were found between the absorbances and the concentrations of the investigated drugs in the concentration ranges of 4–50 and 20–400 μg/mL with TCNQ and pCA, respectively. With all the investigated drugs, TCNQ gave more sensitive assays than pCA; the limits of assay detection were 2.5–4.8 and 20–40 μg/mL with TCNQ and pCA, respectively. The intra- and interassay precisions were satisfactory; the relative standard deviations did not exceed 2%. The proposed procedures were successfully applied to the analysis of the studied drugs in pure form and pharmaceutical formulations with good accuracy; the recovery values were 98.4–102.8 ± 1.24–1.81%. The results obtained from the proposed method were statistically comparable with those obtained from the previously reported methods.

Selective serotonin reuptake inhibitors (SSRIs) have been recently introduced for treatment of depression. The SSRIs are comparable to the tricyclic antidepressants in their clinical efficacy, however, they are more safe and have greater acceptance by the patients. Therefore, SSRIs have become the most widely prescribed antidepressants (1, 2). Five SSRIs are presently available: citalopram, fluoxetine, fluvoxamine, paroxetine, and sertraline; their chemical structures are given in Figure 1. Some SSRIs have also been introduced for other indications, such as obsessive compulsive disorders (3), panic attacks (4), and social phobia (5).

The therapeutic importance of SSRIs was behind the development of numerous methods for their quantitative determination. The methods adapted to the analysis of SSRIs in biological fluids have been reviewed (6). The sophisticated instrumentation of these methods limited their use in quality control laboratories for analysis of these drugs in their pharmaceutical formulations. The methods reported for analysis of 1 or more of the SSRIs in pharmaceutical formulations include titrimetry (7, 8), voltammetry (9–11), polarography (12, 13), capillary electrophoresis (14), and liquid chromatography (LC; 15–20). These methods were associated with some drawbacks, such as being time consuming and/or requiring too expensive instruments that are not available in most quality control laboratories. In spite of the inherent simplicity of spectrophotometry and its availability in almost all the quality control laboratories, the spectrophotometric methods reported for analysis of SSRIs in their pharmaceutical formulations (20–29) suffered from the disadvantages of decreased specificity (20, 21), multiple laborious steps (22–25), and/or long analysis time (26, 27). Moreover, these methods have been developed individually because of the wide differences in the chemical structures of SSRIs. Considering these drawbacks, there was a growing interest in the development of more advantageous spectrophotometric method for determination of SSRIs in their pharmaceutical formulations.

In a previous study (30), a simple and sensitive spectrophotometric method was developed for determination of SSRIs that possess a secondary amine moiety in their structures (fluoxetine, paroxetine, and sertraline). However, the availability of a spectrophotometric method that allows the analysis of all SSRIs drugs would be useful for economic and convenience reasons. Therefore, the aim of the present study was directed to the development of new spectrophotometric procedures that could be applied for analysis of all members of the SSRI group, irrespective of the diversity in their chemical structure. The analytical procedures described herein were based on the formation of colored charge-transfer complexes between the intact molecules of

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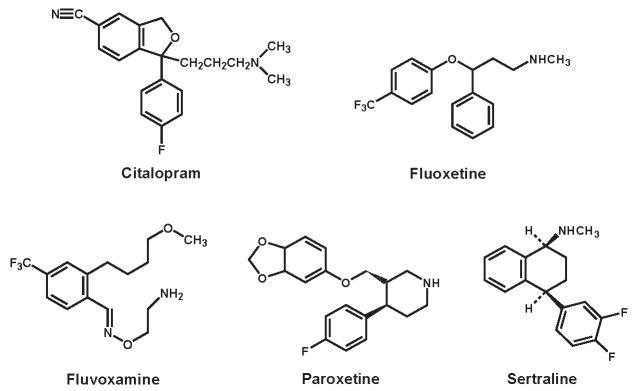

Figure 1. Chemical structures of the SSRIs.

SSRIs as *n*-electron donors and each 7,7,8,8-tetracyanoquinodimethane (TCNQ) or *p*-chloranilic acid (pCA) as π-acceptors. The formed complexes were measured spectrophotometrically at 842 and 520 nm with TCNQ and pCA, respectively.

Experimental

Apparatus

Lambda-3 B (Perkin-Elmer Corp., Norwalk, CT) and UV-1601 PC (Shimadzu, Kyoto, Japan) ultraviolet-visible spectrophotometers with matched 1 cm quartz cells were used for all measurements.

Chemicals and Reagents

Citalopram HBr (Lundbeck, Copenhagen, Denmark); fluoxetine HCl (Hetero Drugs Ltd, Hyderabad, India); fluvoxamine maleate (Solvay Pharma, Suresnes, France); paroxetine HCl (SmithKline Beecham Pharmaceuticals, Brentford, UK); and sertraline HCl (Pfizer Egypt, S.A.E., Cairo, Egypt) were obtained and used as received. TCNQ (Aldrich Chemical Co., Milwaukee, WI) was 0.2% (w/v) in acetonitrile, and the solution was stable for at least 1 week at 4°C. pCA (BDH Chemicals, Poole, UK) was 0.4% (w/v) in acetonitrile; the solution was prepared fresh daily. All solvents and other chemicals used throughout this study were of analytical grade.

Pharmaceutical Formulations

The following commercially available pharmaceutical formulations were used: Cipram tablets (Lundbeck) were labeled to contain 20 mg citalopram HBr/tablet. Prozac capsules (Eli Lilly & Co. Ltd, Hampshire, UK); Philozac capsules (Amoun Pharmaceutical Industries, Cairo, Egypt); Octozac capsules (October Pharma, S.A.E., 6th October, Cairo, Egypt); Fluoxetine capsules (Misr Co. for Pharmaceutical Industries, Cairo, Egypt); Flutin capsules (Egyptian International Pharmaceutical Industries Co., 10th Ramadan, Cairo, Egypt); Florosin capsules (T3A Pharma Group, Assiut, Egypt); and Depreban capsules (Amriya Pharmaceutical Industries, Alexandria, Egypt) were labeled to contain 20 mg fluoxetine HCl/capsule. Faverin tablets (Solvay Pharma) were labeled to contain 50 mg fluvoxamine maleate/tablet. Seroxate tablets (SmithKline Beecham Pharmaceuticals) were labeled to contain 20 mg paroxetine HCl/tablet. Lustral tablets (Pfizer Egypt); Moodapex tablets (Apex Pharma, Cairo, Egypt); and Sirto tablets (Hi Pharma, Cairo, Egypt) were labeled to contain 50 mg sertraline HCl/tablet.

Preparation of Standard and Sample Solutions

(**a**) *Preparation of stock standard solution*.—An accurately weighed amount (400 mg) of each drug was dissolved in 25 mL distilled water in a small beaker. The solution was transferred quantitatively into a 100 mL separating funnel and rendered alkaline with ammonia solution, and the excess ammonia was evaporated by heating the solution in a water bath (Schetzort, Memmert, Germany). The liberated base was extracted with three 25 mL portions of chloroform. The combined extracts were passed through a small funnel containing anhydrous sodium sulfate (2 g) into a 100 mL volumetric flask. The contents of the separating funnel were washed 3 times with chloroform (5 mL for each time). The combined extracts and washings were then diluted to the mark with acetonitrile to obtain stock standard solutions of 4 mg/mL of the investigated drugs calculated as a free base.

(**b**) *Preparation of sample solution of the pharmaceutical formulation*.—Twenty tablets or the contents of 20 capsules of each formulation were weighed and finely powdered. A quantity of the powder equivalent to 400 mg of the active component was transferred into a 50 mL volumetric flask, dissolved in 25 mL of water, swirled and sonicated for 5 min, diluted to volume with water, shaken well for 10 min, and filtered. The first portion of the filtrate was rejected, and a measured volume of the remaining filtrate was transferred quantitatively into a 100 mL separating funnel, and then rendered alkaline with ammonia solution. The procedure was completed as described for preparation of stock standard solution, ensuring that the ratio of chloroform to acetonitrile was the same in both the standard and sample preparations.

Optimization of Experimental Conditions

(**a**) *Concentrations of the acceptor reagents*.—The optimum concentrations of TCNQ and pCA reagents were determined by adding 1 mL of varying concentrations (0.05–0.8%, w/v) to 1 mL of the standard solution (20 and 150 μg/mL for reactions with TCNQ and pCA reagents, respectively). The solutions were diluted to 10 mL with acetonitrile, and the reactions were allowed to proceed at room temperature (25 ± 5°C) for 5 and 20 min for pCA and TCNQ, respectively. The absorbances of the resulting solutions were measured at 520 and 842 nm for pCA and TCNQ, respectively, against reagent blanks treated similarly. The blanks were obtained in a similar manner except 1 mL acetonitrile was added in place of the standard solution.

(**b**) *Solvent*.—The most appropriate solvent for the reaction was determined by carrying out the reaction using the above-mentioned standard concentrations and acceptor

reagent concentrations of 0.2 and 0.4% (w/v, in acetonitrile) for TCNQ and pCA, respectively. The solutions were diluted to 10 mL with acetonitrile, methanol, ethanol, acetone, propan-1-ol, propan-2-ol, *t*-butanol, methylene chloride, chloroform, diethyl ether, toluene, benzene, carbon tetrachloride, 1,4-dioxane, or xylene. The solutions were further treated by the procedures described in (**a**) for optimization of the acceptor concentration.

(**c**) *Reaction time*.—The optimum reaction time was determined by carrying out the reaction in acetonitrile using the above-mentioned standard and acceptor concentrations, and monitoring the color development at room temperature (25 ± 5°C).

General Analytical Procedure

One mL of the standard or sample solution (40–500 and 200–4000 μg/mL for TCNQ and pCA, respectively) was transferred into 10 mL volumetric flasks. One mL of the acceptor solutions (0.2 and 0.4%, w/v, for TCNQ and pCA, respectively) was added. The solutions were then diluted to volume with acetonitrile, and the reactions were allowed to proceed at room temperature (25 ± 5°C) for 5 and 20 min for pCA and TCNQ, respectively. The absorbances of the resulting solutions were measured at 520 and 842 nm in case for pCA and TCNQ, respectively, against reagent blanks treated similarly.

Results and Discussion

All SSRIs, being amines, contain a lone pair of electrons on the nitrogen atom. Therefore, they can potentially act as good *n*-electron donors. The molecular interactions between electron donors and electron acceptors are generally associated with the formation of intensely colored charge-transfer complexes. The rapid formation of these complexes was the basis of development of simple and convenient spectrophotometric methods for these compounds (31–43). For these reasons, a charge-transfer reaction was intended to be involved in the present study. Both TCNQ and pCA are good electron acceptors; however, their interactions with all SSRIs have not yet been studied. Therefore, the present study was designed to evaluate their use in the spectrophotometric analysis of SSRIs via formation of charge-transfer complexes. Because free amines, rather than their corresponding salts, have the electron-donating ability (28, 44), the free bases should be liberated prior to carrying out the reaction. The following sections describe the spectral characteristics of the formed charge-transfer complexes and the optimization of different factors that influence the reaction.

Spectral Characteristics of the Reaction

The interaction of SSRIs with TCNQ and pCA at room temperature gave colored chromogens showing absorption maxima at 842 and 520 nm for TCNQ and pCA, respectively (Figure 2). The predominant chromogen with TCNQ in acetonitrile (polar solvent) is a bluish-green colored radical

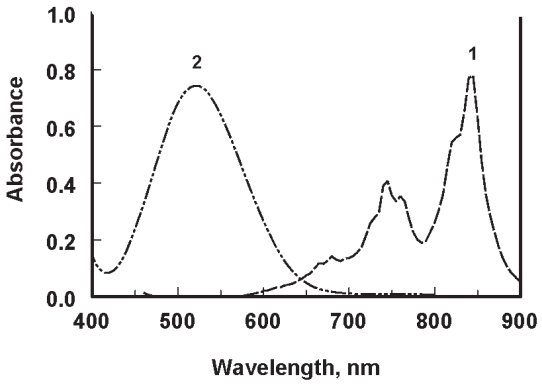

Figure 2. Absorption spectra of the reaction products of fluoxetine with TCNQ (1) and pCA (2). The final concentrations of fluoxetine were 20 and 150 μg/mL for TCNQ and pCA, respectively.

anion, which exhibits strong absorption maxima at 842, 825, 762, and 742 nm. These bands may be attributed to the formation of the radical anion $TCNQ^-$, which was probably formed by the dissociation of an original donor-acceptor (D-A) complex with SSRIs. The dissociation of the complex was promoted by the high ionizing power of acetonitrile.

$$D + A \rightleftharpoons \underset{\text{complex}}{(D\text{-}A)} \overset{\text{polar solvent}}{\rightleftharpoons} \underset{\text{radical ions}}{D^{+} + A^{-}}$$

Further support of this assignment was provided by comparing the absorption maxima with those of TCNQ radical anion produced by the iodide reduction method (45). The dissociation of the D-A complex was promoted by the high ionizing power of the polar solvent, and the resulting peaks in the absorption spectra of SSRI-acceptor reaction mixtures were similar to the maxima of the radical anions of the acceptors obtained by the iodide reduction method (45).

pCA exists in 3 ionic forms: neutral yellow-orange H_2A at very low pH, dark purple HA^- that is stable at pH = 3, and a pale violet A^{2-} that is stable at high pH. These transformations are illustrated in the following scheme:

$$H_2A \rightleftharpoons H^+ + HA \text{ (violet)}$$
$$HA \rightleftharpoons H^+ + A^{2-} \text{ (colorless)}$$

Because the interaction of SSRIs with pCA in acetonitrile gave a violet product, it might be concluded that HA^- was the form of pCA involved in the reaction described herein.

Optimization of Reaction Conditions

The results of variations in the reagent concentrations indicated that 1 mL of 0.2 and 0.4% (w/v) TCNQ and pCA, respectively, were optimum whereas the highest absorbances were obtained. In order to select the most appropriate solvent, the reactions were carried out in different solvents. Small

Table 1. Effect of solvents on the position and intensity of absorption of the charge-transfer complexes formed from the reaction of SSRIs with TCNQ and pCA[a]

		TCNQ		pCA	
Solvent	Dielectric constant[b]	λ_{max}, nm	ε_{max}	λ_{max}, nm	ε_{max}
Acetonitrile	37.5	842	12013	520	1600
Methanol	32.7	852	10775	528	1342
Ethanol	24.6	847	8775	525	1410
Acetone	20.7	846	10721	523	1139
Propan-1-ol	20.3	848	8742	525	1135
Propan-2-ol	19.9	852	8418	530	1092
Butan-1-ol	17.5	848	8207	525	1064
t-Butanol	12.5	854	7625	530	1098
Methylene chloride	8.9	851	9041	530	1020
Chloroform	4.8	842	8079	520	1040
Diethyl ether	4.3	852	6650	530	863
Toluene	2.4	855	6390	536	830
Benzene	2.3	855	7299	533	945
Carbon tetrachloride	2.2	843	6747	520	876
1,4-Dioxane	2.2	842	5855	520	759
Xylene	2.0	842	5855	520	759

[a] Using fluoxetine as a representative example; the concentrations were 20 and 150 mg/mL for reaction with TCNQ and pCA, respectively.
[b] Values were obtained from ref. 46.

shifts in the position of the maximum absorption peak were observed, and the molar absorptivities were also influenced (Table 1). The data indicated that the interaction of SSRIs with TCNQ and pCA in polar solvents (e.g., acetonitrile) produced charge-transfer complexes with molar absorptivity (ε) values higher than those produced in nonpolar solvents (e.g., chloroform). The values of ε were correlated with the dielectric constants (46) of the solvent in which the reaction was performed (Figure 3); $r^2 = 0.8030$ and 0.8734 for TCNQ and pCA, respectively. This was attributed to the complete electron transfer from the SSRIs (electron donor) to TCNQ and pCA (electron acceptor) that takes place in the polar solvents (e.g., acetonitrile and methanol). Acetonitrile was selected for the subsequent experiments because it offered maximum sensitivities with both TCNQ and pCA. This was attributed to the high dielectric constant of acetonitrile, which promotes maximum yield of radical anions, in addition to its high solvating power for the acceptors.

The optimum reaction time was determined by monitoring the color development at room temperature (25 ± 5°C). Complete color development was attained instantaneously with pCA and after 20 min with TCNQ. In spite of the instantaneous reaction with pCA, however, for more precise readings, the reaction solution was allowed to stand for 5 min before measurements. The developed colors with both acceptors remained stable at room temperature for at least another 30 min.

Validation of the Proposed Methods

Linearity and sensitivity.—Under the above-mentioned optimum reaction conditions, the calibration graphs for the analysis of SSRIs by TCNQ and pCA were constructed by plotting the absorbances as a function of the corresponding concentrations. The regression equations for the results were derived using the least-squares method. In all cases, Beer's law plots (n = 5) were linear with very small intercepts (0.0056 – 0.0251) and good correlation coefficients

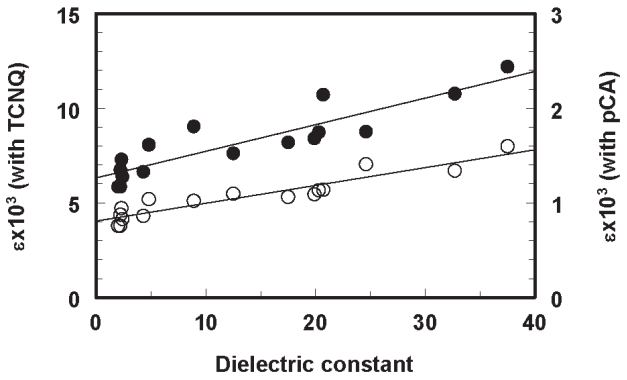

Figure 3. Correlation of the dielectric constants of the solvents with the molar absorptivities (ε) of the charge-transfer complexes of SSRIs with TCNQ (●) and pCA (○).

Table 2. Quantitative parameters for the spectrophotometric analysis of SSRIs by formation of charge-transfer complexes with TCNQ and pCA

Compound	Range, μg/mL	Intercept (a)	Slope (b)	Correlation coefficient (r)	Molar absorptivity, L/mol cm	LOD, μg/mL	LOQ, μg/mL
			TCNQ				
Citalopram	5–50	0.0080	0.0192	0.9975	6490	4.8	14.4
Fluoxetine	4–25	0.0011	0.0380	0.9996	11750	2.5	7.5
Fluvoxamine	5–40	0.0056	0.0241	0.9985	7640	4.0	12.0
Paroxetine	4–32	0.0077	0.0300	0.9992	9880	3.0	9.0
Sertraline	4–30	0.0108	0.0323	0.9986	10110	2.8	8.4
			pCA				
Citalopram	40–400	0.0067	0.0024	0.9994	810	40	120
Fluoxetine	20–200	0.0142	0.0048	0.9987	1500	20	60
Fluvoxamine	30–300	0.0050	0.0032	0.9996	1030	30	90
Paroxetine	25–280	0.0146	0.0034	0.9992	1140	25	75
Sertraline	20–240	0.0251	0.0039	0.9986	1220	20	60

(0.9975 – 0.9996) in the concentration ranges of 4–50 and 20–400 μg/mL in TCNQ and pCA, respectively (Table 2). The limits of detection (LOD) and quantitation (LOQ) were determined using the formula: LOD or LOQ = κSDa/b, where κ = 3 for LOD and 10 for LOQ, SDa is the standard deviation of the intercept, and b is the slope. The LOD and LOQ values obtained using TCNQ were 2.5–4.8 and 7.5–14.4 μg/mL, respectively. When using pCA, these values were 20–40 and 60–120 μg/mL, respectively. In all cases, TCNQ gave higher sensitivities (greater ε values) than pCA. This might be attributed to the greater electron affinity of TCNQ. With each particular acceptor, the ε values for secondary amines (fluoxetine, paroxetine, and sertraline) were higher than that for the primary amine (fluvoxamine), which in turn was higher than that for the tertiary amine (citalopram). These data were coincident with the reported relative basicities (electron-donating abilities) of the amines (47).

Accuracy, precision, and specificity.—Analytical recovery for the proposed procedures was determined by the standard addition method. Known amounts of SSRI (20 and 150 μg/mL for TCNQ and pCA, respectively) were added to predetermined SSRIs containing dosage forms and then determined by the proposed methods. The mean analytical recovery was calculated as the ratio between the concentrations measured to the concentrations taken for analysis, expressed as percentages. The mean analytical recovery for the proposed procedures ranged from 98.4–102.8 ± 0.69–1.76% (Table 3), indicating the accuracy of the method. The precision of the assays were determined on SSRI solutions containing 20 and 150 μg/mL of each compound for the TCNQ and pCA procedures, respectively. The intra-assay precision was assessed by analyzing 6 replicates of each sample as a batch in a single assay run, and the interassay precision was assessed by analyzing the same sample in triplicate in 2 separate assay runs on 2 consecutive days. The obtained results were satisfactory; the relative standard deviation (RSD)values were less than 2% (Table 4). This level of precision is adequate for the routine analysis of SSRIs in quality control laboratories. The measurements in the proposed spectrophotometric procedures are carried out in the visible region, thus avoiding interferences from the ultraviolet (UV) absorbing interfering substances that might be coextracted from the dosage forms. Furthermore, the satisfactory results of the recovery studies proved the absence of interferences from the excipients of the dosage forms with the proposed reactions.

Ruggedness and robustness.—The ruggedness of the proposed methods was assessed by applying the procedures using 2 different instruments (cited under *Apparatus*) in

Table 3. Analytical recoveries for the spectrophotometric analysis of SSRIs by TCNQ and pCA

	Recovery, % ± SD[b]	
Compound[a]	TCNQ	pCA
Citalopram	102.8 ± 1.48	101.4 ± 1.47
Fluoxetine	100.5 ± 1.27	98.4 ± 1.76
Fluvoxamine	98.9 ± 1.29	102.0 ± 1.51
Paroxetine	101.6 ± 0.84	99.9 ± 0.69
Sertraline	99.6 ± 1.45	100.4 ± 1.26

[a] Concentrations of SSRIs were 20 and 150 μg/mL for TCNQ and pCA, respectively.

[b] Values are mean of 5 determinations; SD = standard deviation.

Table 4. Precision of the proposed methods for analysis of SSRIs by TCNQ and pCA

	Within-assay, n = 6		Between-assays, n = 6	
Compound[a]	TCNQ	pCA	TCNQ	pCA
Citalopram	19.98 (1.28)[b]	152.02 (1.45)	20.52 (1.92)	147.98 (1.58)
Fluoxetine	19.78 (1.07)	148.42 (1.59)	19.35 (1.89)	149.78 (1.87)
Fluvoxamine	20.53 (0.89)	150.67 (1.11)	20.55 (1.70)	150.53 (1.59)
Paroxetine	20.87 (1.06)	147.28 (0.95)	18.98 (1.89)	153.87 (1.96)
Sertraline	19.06 (1.12)	148.52 (1.28)	19.68 (1.45)	147.06 (1.92)

[a] Concentrations of SSRIs were 20 and 150 μg/mL for TCNQ and pCA, respectively.

[b] Values are the measured concentrations in μg/mL. Values in parentheses are the RSD values (%).

2 different laboratories at different elapsed times. The RSD values for the results were calculated. Robustness of the procedures was assessed by evaluating the influence of small variation in the experimental variables: concentrations of acceptor (0.2–0.3 and 0.3–0.5%, w/v, for TCNQ and pCA, respectively) and reaction time (optimum ± 2 min) on the analytical performance of the method. In these experiments, 1 experimental parameter was changed while the other parameters were kept unchanged, and the recovery was calculated each time. Laboratory-to-laboratory, day-to-day, and small variations in the experimental variables did not significantly affect the analysis performance. The recoveries were 98.3–102.5 ± 0.75–1.84%, and the results were found to be reproducible because the RSD did not exceed 2%. This indicated the reliability of the proposed methods during routine work.

Analysis of the Pharmaceutical Formulations

The commercially available pharmaceutical formulations of the investigated compounds were subjected to the analysis by the proposed and previously reported methods (16, 20, 22, 29, 48), and the obtained results were then statistically compared with each other. The mean recoveries, relative to the labeled amounts, obtained by the proposed methods were 98.4–101.8 ± 1.24–1.81% (Table 5). In the t- and F-tests, no significant differences were found between the calculated and

Table 5. Analysis of SSRIs in their pharmaceutical formulations by the proposed spectrophotometric and previously reported methods

	Content, % ± SD[a]		
Pharmaceutical formulation	TCNQ	pCA	Reported[b]
Cipram tablets	99.2 ± 1.24 (2.12, 2.27)[c]	98.4 ± 1.47 (2.45, 4.54)	98.9 ± 0.69[b]
Prozac capsules	99.6 ± 1.92 (2.51, 2.36)	99.7 ± 1.72 (2.29, 1.89)	100.3 ± 1.25
Philozac capsules	99.1 ± 1.58 (2.28, 1.99)	99.5 ± 1.81 (0.62, 2.61)	99.6 ± 1.12
Octozac capsules	99.2 ± 1.84 (1.60, 2.27)	100.1 ± 1.68 (2.13, 1.90)	99.6 ± 1.22
Fluoxetine capsules	100.6 ± 1.58 (2.19, 1.60)	99.8 ± 1.58 (1.32, 1.60)	100.1 ± 1.25
Flutin capsules	99.3 ± 1.48 (2.49, 2.28)	100.2 ± 1.54 (1.94, 2.47)	99.8 ± 0.98
Florosin capsules	100.2 ± 1.63 (2.04, 1.30)	99.6 ± 1.81 (0.38, 1.60)	99.7 ± 1.43
Faverin tablets	100.7 ± 1.63 (2.36, 1.12)	101.8 ± 1.59 (2.00, 1.07)	101.3 ± 1.54
Depreban capsules	99.8 ± 1.28 (2.30, 2.27)	99.8 ± 1.84 (1.74, 4.69)	100.2 ± 0.85
Lustral tablets	100.6 ± 1.55 (1.96, 2.78)	99.9 ± 1.52 (1.49, 2.67)	100.2 ± 0.93
Moodapex tablets	99.2 ± 1.64 (1.44, 3.72)	99.1 ± 1.48 (2.07, 3.03)	99.5 ± 0.85
Sirto tablets	100.7 ± 1.33 (2.43, 1.15)	100.8 ± 1.65 (1.71, 1.77)	101.2 ± 1.24
Seroxate tablets	100.6 ± 1.62 (2.42, 3.63)	100.7 ± 1.62 (1.93, 3.63)	101.1 ± 0.85

[a] Values are mean of 5 determinations.

[b] Refs. 16, 20, 22, 29, and 48 for paroxetine, sertraline, fluvoxamine, fluoxetine, and citalopram, respectively.

[c] Values in parentheses are the calculated values of t and F; the tabulated values at the 95% confidence limit are 2.78 and 6.39, respectively.

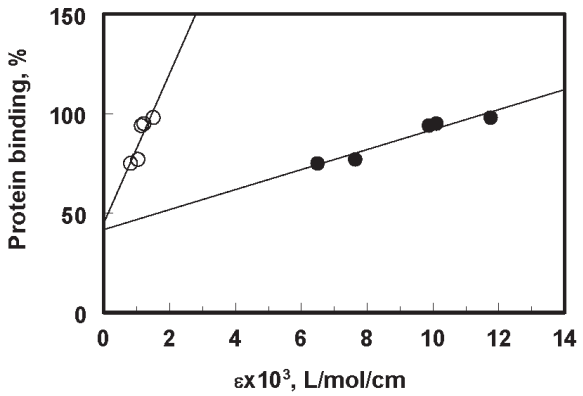

Figure 4. Correlation of the protein binding of SSRIs with the experimental molar absorptivities (ε) of their charge-transfer complexes with TCNQ (●) and pCA (○).

theoretical values of both the proposed and previously reported methods at the 95% confidence level. This indicated similar accuracy and precision in the analysis by the proposed and previously reported methods.

Correlation Between Molar Absorptivity and Protein Binding

The pharmacokinetics and pharmacodynamics of SSRIs are influenced by their binding to the body proteins (1). Therefore, the possible role of electron transfer in drug-protein binding was studied; TCNQ and pCA were used as models for electron acceptors. The sensitivities, expressed as ε values, for the reaction of SSRIs with both acceptors were found to be dependent on the average percent protein binding of the investigated SSRIs (Figure 4). The regression equations for the correlation between ε values and percentage of protein binding were:

$$\% \text{ Protein binding} = 41.71 + 0.01\ \varepsilon \quad (r^2 = 0.9346) \text{ for TCNQ}$$

$$\% \text{ Protein binding} = 44.95 + 0.04\ \varepsilon \quad (r^2 = 0.7604) \text{ for pCA}$$

TCNQ gave a more sensitive assay and better correlation with protein binding.

Conclusions

The proposed methods are superior to most of the previously reported spectrophotometric methods for any of the SSRIs in terms of greater sensitivity (27), simpler assay procedures (22–25), and shorter analysis time (26, 27). Moreover, the proposed methods, rather than any of the previously reported methods, could be used for the analysis of all SSRI drugs, irrespective of the diversity in chemical structure. These advantages give the proposed methods great value in quality control analysis of SSRIs in their pharmaceutical formulations.

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