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A Novel Clomipramine and Paroxetine-Selective Membrane Sensors and their Applications in Pharmaceutical Analysis

Hoda Mahmoud Elqudaby[1], Eman Yossri Zaki Frag[2], Gehad Genidy Mohamed[2,*] and Mohamed Abd El-Razek Mohamed[2]

[1]*National Organization for Drug Control and Research, Elharam St., Giza, Egypt*
[2] *Chemistry Departments, Faculty of Science, Cairo University, 12613 Giza, Egypt*

* Corresponding Author, Tel: 002-01523423103; Fax: 002-02-35728843

E-Mail: ggenidy@hotmail.com

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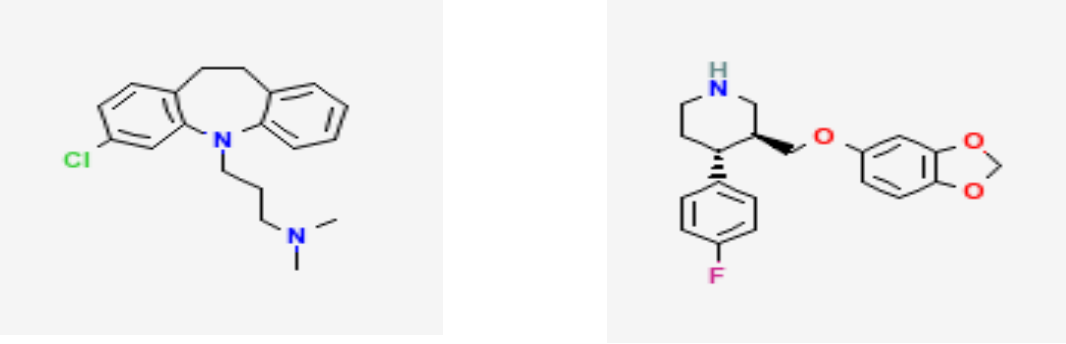
Abstract- Selective electrodes based on the reineckate drugs ion pairs in a polyvinyl chloride (PVC) matrix membrane is proposed for the determination of clomipramine (CLO) and paroxetine (PRX) hydrochlorides. The ion-pair complexes of drug cations with reineckate anion are either dissolved in tetrahydrofuran solvent or dispersed in a PVC matrix, with dioctylphthalate or dioctyl sebacat plasticizers, and used as the ion-exchange membranes. The electrochemical response characteristics of electrodes incorporating these types of membranes are evaluated with regard to the effect of pH, foreign basic compounds, effect of soaking time and lifespan. A well defined internal electrode potential was achieved using Ag/AgCl disc in contact with the ion-pair electroactive membrane phase. The electrodes display a stable fast Nernstian response in the concentration ranges 10^{-2}-10^{-7} mol L^{-1} for the two drug cations over the pH range 2-5, a short static response time less than 60 s and response remain stable for at least 6 weeks. The lower limit of detection was 10^{-7} mol L^{-1} for both drugs. Determination of as low as 8 μg mL^{-1} of drugs hydrochloride shows an average recovery of 98.75 and 100.6% and a mean standard deviation of 0.70 and 0.35 for CLO and PRX drugs, respectively. Selectivity coefficients for CLO and PRX drugs relative to a numbers of potential interfering substances were investigated. The electrodes exhibit useful analytical characteristics for determining CLO and PRX in some illicit powders. The results agree fairly well with those obtained by gas-liquid chromatography. The proposed electrodes are simple easy to construct and behave as symmetric ion selective electrodes (ISEs).

Keywords- Ion Selective Electrode, PVC Membrane, Clomipramine, Paroxetine

1. INTRODUCTION

The ion selective membrane electrode technique has become a satisfactory tool for pharmaceutical analysis, although no pharmacopoeia has introduced their use for analysis. Some pharmaceutical analysis introduced membrane electrodes which enable the activities of various drugs to be measured directly and selectively [1-5], and in most cases, without prior separation of the tested drug from the formulations matrix.

Clomipramine hydrochloride (Fig. 1) is 3-chloro-5-[3-(dimethylamino) propyl]-10, 11-dihydro-5H-dibenz-[b, f] azepine monohydro-chloride and paroxetine (Fig. 1) is (3S, 4R)-3-[(1,3-benzodioxol-5-vloxy)methyl-[4-(4-flurophenyl)piperidine are typically antidepressant drugs, they are widely used in the treatment of mood disorders, particularly depression and anxiety disorders**.**



CLO PRX

Fig. 1. Structure of clomipramine (CLO) and paroxetine (PRX) drugs

Several spectrophotometric methods have been reported for the determination of clomipramine hydrochloride [6-10]. It could also be determined electrochemically using different techniques [11-13]. Chromatographic techniques were suggested for the determination of the drug in row materials and in biological fluids using different detectors [14-18]. Fast LC-MS/MS [19], capillary gas chromatographic with flame ionization detection [20] as well as capillary zone electrophoresis methods [20-22] were also used Paroxetine was determined by different methods such as chromatographic [23-29] and electrochemical methods [30,31]. Paroxetine was determined spectrophotometrically via charge transfer complex formation with TCNQ and other reagents [32-34]. Potentiometric detection based on ion-selective electrodes (ISEs), offers several advantages such as speed and ease of preparation and procedures, simple instrumentation, relatively fast response, wide dynamic range, reasonable selectivity, and low cost [19,20]. Based of our knowledge, there is no previous reports regarding the determination of clomipramine (CLO) and paroxetine (PRX) based on ISE. In this paper, we introduced new potentiometric sensors for selective

determination of clomipramine (CLO) and paroxetine (PRX) drugs in pharmaceutical compounds. The method is based on the ion-pair formation between clomipramine (CLO) and paroxetine (PRX) drugs and ammonium reinekate as an electroactive material and dioctyl phthalate (DOP) and dioctyl sebacate (DOS) as anion excluders in PVC matrix. Electrodes with reineckate ion pair complexes in polyvinyl chloride membranes have been prepared, characterized, compared and examined for drugs determination. These electrodes display stable, fast and linear response for 10^{-2}-10^{-7} mol L^{-1} CLO and PRX hydrochlorides over the pH range 2-5 with minimum interference from many related basic compounds.

2. EXPRIMENTAL

2.1. Reagents

All reagents and chemicals used were of analytical reagent grade and solutions were prepared with doubly distilled water. Ammonium reineckate reagent of purity 98% were obtained from Sigma (St. Louis, MO, USA). Polyvinyl chloride (PVC), dioctylphthalate (DOP), dioctyl sebacat (DOS) and tetrahydrofuran (THF) were obtained from Aldrich Chem. Comp., Inc., and Milwaukee, WI, USA. CLO row material was supplied from Acapi Co., Egypt. Anapramine (75 mg/tablet) was supplied from Sigma pharmaceutical industries, Egypt. Anafranil (25 mg/tablet) was supplied from Novartis, 10 Ramadan city, Egypt. PRX row material was supplied from Elpheronea Co, El-Obor city, Egypt. Paroxetine (20 mg /tablet) was supplied by Eva pharma, Egypt. Xanadole (20 mg/tablet) was supplied by European Egyptian Pharm, El-Ameria city, Egypt.

0.1 mol L^{-1} solutions of the drugs under investigation were prepared by dissolving the proper weight of each drug into suitable amount of 0.002 mol L^{-1} HCl which was added dropwise with continuous stirring till complete drug dissolution was achieved. Working solutions of each drug in concentration range 10^{-7}-10^{-2} mol L^{-1} for CLO and PRX were prepared by serial accurate dilution in acetate buffer of pH 4.50. The stock solutions and the dilutions were kept in dark bottles in the refrigerator. 10^{-2} mol L^{-1} Ammonium reineckate ($NH_4Cr(SCN)_4(NH_3)_2H_2O$), standard solution was prepared by dissolving 0.354 g into 50 ml of doubly distilled water.

2.2. Apparatus

All potentiometric measurements were carried out at 25±2 ºC using an Orion pH/mV meter (Model SA 720) with pH sensitivity of ±0.05 pH units with drug-PVC membrane electrodes in conjunction with an Orion Ag/AgCl double junction reference electrode (Model 90-02) with 10% w/v potassium nitrate in the outer compartment. An Orion combination pH electrode (Model 91-02) was used for pH adjustments.

2.3. Reineckate drug ion- pair complexes

The ion pair complexes of the cited drugs were prepared by mixing equal volume of 1×10^{-2} mol L^{-1} aqueous ammonium reineckate and 1×10^{-2} mol L^{-1} drug (CLO and PRX) and they were added dropwise with continuous stirring. The resulting precipitates were left in contact with their mother liquor over night to ensure complete coagulation, then filtered off through a Whatman filter paper No. 42 and washed thoroughly with distilled water several times and left to dry at room temperature for at least 24 h and ground to fine powder [35].

For preparation of the membrane, 5.0 mL of 1.0×10^{-2} mol L^{-1} aqueous clomipramine (CLO) and paroxetine (PRX) drugs solution was added to 10 mL of 1.0×10^{-2} mol L^{-1} solution of ammonium reineckate. The resulting precipitate was filtered off through a Whatman filter paper No.42 and washed with deionized water and dried, protected from light in a desiccator at room temperature. Then, about 10.0 mg of this precipitate was mixed with 190 mg PVC and 350 mg of DOS or DOP plasticizers previously dissolved in 5 mL of THF. The resulting homogeneous mixture was then poured into a 20 mm Petri dish, covered with a filter paper and the solvent was allowed to evaporate at room temperature. Semi-transparent PVC membrane was obtained with an average thickness of about 0.2 mm. A Pyrex tube (3 mm i.d.) was dipped into the mixture for about 10 s so that a membrane was formed. Then, the tube was then pulled out from the mixture and kept at room temperature for 12 h. The tube was then filled with the internal solution (1.0×10^{-2} mol L^{-1} clomipramine (CLO) and paroxetine (PRX)). The filled electrode was conditioned by soaking into 1.0×10^{-4} mol L^{-1} clomipramine (CLO) and paroxetine (PRX). The first conditioning time was approximately 24 h and then was about 30 min for successive uses. An Ag/AgCl electrode was used as an internal reference electrode. The PVC membrane electrodes were conditioned by soaking in 1.0×10^{-4} mol L^{-1} clomipramine (CLO) and paroxetine (PRX) solution for 1 day before measurements and stored in air when not in use [36-39].

2.4. Procedures

2.4.1. Effect of pH and response time

The effect of pH of the test solution on the potential values of the electrode system in solutions of different concentrations of CLO and PRX drugs was tested using two concentrations of each drug (10^{-3} and 10^{-4} mol L^{-1}) over a range of pH (2-11) and recording the potential readings of the studied sensors by immersing the corresponding drug-sensor and a double junction Ag/AgCl reference electrode in 50 ml beakers containing 25 ml aliquots of dilutes of 10^{-3} and/or 10^{-4} mol L^{-1} drug-aqueous solutions. The pH of each solution was gradually changed by adding small aliquots of dilute sodium hydroxide and/ or hydrochloric acid solutions. The mV versus pH profile of each drug concentration was plotted for each electrode system. The potential readings that were insensitive to pH changes were obtained

from the mV–pH plots. The dynamic response time of the studied sensors were tested by measuring the time required to reach a steady potential within ±0.3 mV.

2.4.2. Effect of soaking time and life span of the electrodes

Each of the investigated electrodes was soaked in 10^{-2} mol L^{-1} solution of the corresponding drug at 25 ^{o}C. The potential response was measured for each electrode after time intervals covering the range of 24 h up to two months. The measurements were stopped when the slope of the calibration curve deviated largely from the Nernstian value and the electrode become out of use. Also, sensor life span of each sensor was examined by repeated monitoring of the slope of each separate sensor. In order to regenerate the exhausted electrodes containing individual ion-exchanger, there were soaked in 10^{-2} mol L^{-1} RK at 25 ^{o}C for 24 h followed by soaking in the studied drug (10^{-2} mol L^{-1}) solution for several hours depending on the electrode type.

2.4.3. Potentiometric titrations

Different aliquots of the investigated drugs (10^{-2} mol L^{-1}) have been transferred into 50 ml titration cell and diluted to 10 ml by acetate buffer of pH 4.5. The resulting solutions were stirred and titrated against 10^{-2} mol L^{-1} RK according to the corresponding ion-exchanger used in constructing the electrode. The electrode potential (E) was recorded against the volume of the titrant added (V) and plotted as E *vs*. V curve and the end point was determined by the conventional S-shaped curves [40].

2.4.4. Effect of plasticizers

Polyvinyl chloride (PVC) membranes plasticized with different plasticizer as dioctyl sebacat (DOS) or dioctylphthalate (DOP) and prepare the corresponding membrane for each drug and calculate the slope of the corresponding calibration graphs at 25±1^{o}C. Potentiometric response of the proposed electrodes for the investigated drugs based neutral ionophores is greatly influenced by the polarity of the membrane medium, which is in turn defined by the dielectric constants of the major membrane incorporating (drug) with different solvents.

2.4.5. Determination of sensor selectivity

The influence of some inorganic cations, sugars and as additives on the response of the CLO-RK and PRX-RK electrodes towards their respective drugs were tested using the separate solution method [41], which requires two potential measurements. First, the potential is measured in a solution containing a known activity of the ion for which the electrode is selective. Second, the potential in a solution containing the interfering ion, was used to determine the selectivity coefficient value ($K_{d,i}$), where (d) is drug primary ion and (i) interfering secondary ion solutions of equal concentrations were prepared. The potentials E_d and E_i were measured using prepared CLO-RK and PRX-RK ISEs in the cell. Standard

solutions (10^{-3} mol L^{-1}), of Na(I), K(I), Ca(II), Mn(II), Ba(II), Co(II), Mg(II), Cu(II), Zn(II), urea, starch, glucose, lactose, sucrose, fructose, CLO and PRX are used as interfering ions for the selectivity study and the electrode signal (mV), for each interfering ion is recorded. A 1.0 ml aliquot of 10^{-3} mol L^{-1} of the drug solution was transferred into 50 ml beaker containing 9.0 ml of acetate buffer of pH 4.5. The drug sensors in conjunction with Ag/AgCl reference electrode were immersed in the solution and the potential reading was measured (E_{Drug}). In a separate run, a 1.0 ml aliquot of 10^{-3} mol L^{-1} of the interfering solution was transferred into a 50 ml beaker containing 9.0 ml of the same buffer and the potential reading was recorded (E_i) and selectivity coefficients were calculated [41].

2.4.6. Calibration curves

After soaking the electrode in 10^{-4} mol L^{-1} solution of the corresponding drug, it immersed in acetate buffer for 30 min then aliquots of (10 ml) each drug sample solutions in acetate buffer of pH 4.50 having concentrations of 10^{-7}-10^{-2} mol L^{-1} for each drugs were transferred into 50 ml beakers. The potential in mV of each sample solution was directly measured using the respective ISEs (drug-reineckate ion pair complex) based on DOS or DOP as plasticizer as mentioned in membrane preparation. The working electrode in conjunction with a double junction Ag/AgCl reference electrode was immersed in the drugs solution. The emf readings were recorded after stabilization to 0.5 mV and plotted *vs.* log [drug. hydrochloride]. The calibration graph was constructed and used for subsequent measurement of drugs in illicit powders.

2.4.7. Direct determination of clomipramine (CLO) and paroxetine (PRX)

Five tablets of the drug formulations were weighed and finely powdered in a small dish. An accurately weighed portion of the powder equivalent to 1.929 g PRX and 0.485 g CLO was dissolved in the minimum volume of distilled water and few drops of 0.002 M HCl by sonication for 10 min. The solution mixture was shaken in a mechanical shaker and accurately transferred to 50 ml measuring flask. The pH of the solution was then adjusted to pH 5 with acetate buffer of pH 4.5 and shaken well to form 1×10^{-2} mol L^{-1} solutions. 10, 15, and 20 ml aliquots of each drug solution were transferred into 25 ml volumetric flasks and made up to the mark with acetate buffer of pH=4.5. Solutions having concentrations of 4×10^{-3}, 6×10^{-3} and 8×10^{-3} mol L^{-1} of CLO and PRX were produced. Potentials of the resulting drug solutions were directly measured using its corresponding ion selective electrode.

2.4.8. Standard addition method

Alternatively, the standard addition (spiking) method was used as follows: transfer a known volume of the test solution (1–8 ml) of the drug at pH 4.5 into a 25 mL measuring flask. Measure the potentials displayed by the test solution of the drug before and after

addition of 1ml aliquot of standard CLO and PRX solution (1×10^{-2} mol L^{-1}). The change in the electrode potential (E) was then recorded and used for determining the drug.

3. RESULTS AND DISCUSSION

Ion selective electrodes are a technology that gives direct measurement of these drugs. Time consuming steps such as filtrations, weightings and distillation are not required in most cases. Concentrations can be read out directly on a pH/mV/ION meter, or can be read from a constructed calibration curve. ISEs presented here are based on polyvinylchloride (PVC) membrane supported with electro-active ion pair complexes. Electrode methods save time, and since electrodes are portable, measurements can be made on a laboratory bench, the bank of a river or pond, the floor of a manufacturing plant, or the tray of a truck .

Clomipramine (CLO) and paroxetine (PRX) hydrochlorides behave as cations in acidic medium, due to presence of the amino groups. This fact suggests the use of anionic type of ion exchangers, DOP, DOS and ammonium reinekate with their low solubility products and suitable grain size.

The PVC was used as a polymer matrix in fabrication of membrane sensors. Clomipramine (CLO) and paroxetine (PRX) hydrochlorides were found to form 1:1 ion association complexes with each ammonium reinekate as proved by elemental analysis data. The elemental analysis of the formed CLO-RK and PRX-RK ion pairs is carried out and the results obtained are: %Found (calculated): C = 41.80 (39.14), H = 4.22 (4.80), N = 16.75 (17.80) and C = 38.30 (37.87), H = 4.76 (4.30) and N = 16.22 (15.36) for CLO-RK and PRX-RK ion pairs, respectively.The obtained data for the determination of CLO and PRX drugs using DOS or DOP as plasticizers were very close to each other and the two PVC membrane electrodes give high sensitivity and selectivity for the determination of CLO and PRX drugs at very low concentrations of 10^{-7}-10^{-2} mol L^{-1}. The preformed electrodes can be used for at least 5 and 6 weeks for CLO and PRX, respectively, with very fast response to the drugs under investigation. To the drugs under investigation. Potentiometric response characteristics for CLO-RK and PRX-RK membrane electrodes are given in Table 1.

3.1. Effect of plasticizer type

A membrane is a phase and is finite in space which separates two other phases and exhibits individual resistance to the permeation of different species. Polymer matrix provides mechanical stability of the membrane, chemical stability, clean surface of the resulting membrane, chemical inertness and can be adjusted to exhibit extra requirements, i.e., physiological fluids sample, biocompatibility, adhesion, etc. As the results showed, among the two different plasticizers used, DOP and DOS were more effective solvent mediator in preparing the CLO and PRX membrane sensors. It should be noted that the nature of the plasticizer influences both the dielectric constant of the membrane and the mobility of the

ionophore and its complex. Initially, plasticizers were applied to the polymer matrix in order to decrease its viscosity and provide mobility of the membrane constituents within the membrane phase. In relation to the role of plasticizer in a PVC membrane, it should be noted that plasticizer acts as a membrane solvent, affecting membrane selectivity through both extraction of ions into the organic phase. Both membrane solvents of low dielectric constants ε (adipates, sebacates and phthalates, ε=∼4) are available [42]. The critical response characteristic of the proposed electrodes was investigated according to IUPAC recommendations [42]. Different membrane composition was checked using DOS and DOP as plasticizers.

Table 1. Potentiometric response characteristics for CLO-RK and PRX-RK membrane

Parameter	CLO- RK sensor		PRX- RK sensor	
	DOS	DOP	DOS	DOP
Working concentration range, (mol L^{-1})	10^{-7}–10^{-2}	10^{-7}–10^{-2}	10^{-7} –10^{-2}	10^{-7}–10^{-2}
Slope, mV $decade^{-1}$	52.28 0.15	52.00±0.21	51.00±0.3	50.40±0.
Intercept	380.29	392.00	368.3	32
Lower limit of detection, (mol L^{-1})	1×10^{-7}	1×10^{-7}	1×10^{-7}	283.5
Correlation coefficient (r)	0.995	0.995	0.9998	1×10^{-7}
Working pH range	4.5	4.5	4.5	0.9950
Response time, (sec)	20	25	20	4.5
Life span, (week)	6	6	5	25
				5

3.2. Effect of pH

The influence of pH on the response of the proposed drug selective electrodes was checked by measuring the potential displayed by 10^{-3} and 10^{-4} mol L^{-1} drug test solutions over the pH range 2-11 (Fig. 2). Small volumes of dilute sodium hydroxide and/ or HCl were used for adjustment. The responses of the two sensors are fairly constant in acetate buffer solutions of pH 2-5. The potential did not vary over this range by more than ±5 mV and the electrode can be applied for the determination of the investigated drugs. A considerable decrease of the potential above pH 6 was observed and it is probably due to the decreased concentration of the protonated form of drug whereas at higher pH, the disturbances happen in the potentials reading due to the penetration of the hydronium ion into the membrane gel layer [43].

3.3. Effect of soaking time, lifespan and response time of the electrodes

In this work, the performance characteristics of the investigated electrodes were studied as a function of soaking time. For this purpose, the electrodes were soaked in solution (10^{-4} mol L^{-1}) of the respective drug for different time intervals starting from 1 h to reaching 42 days. The slope of the calibration graph was found to be 51.00±0.96 and 52.28±1.09 mV $decade^{-1}$ after 24 h of soaking and then it starts to decrease gradually till reaches about 49.4±0.46 and 45±0.69 mV $decade^{-1}$ after 35 and 42 days of continuous soaking for PRX-RK and CLO-RK sensors, respectively. On the other hand, continuous soaking of the electrodes in 10^{-4} mol L^{-1} solution of the drug affected negatively their response to drug cation. This negative effect of soaking is attributed to the leaching of the active ingredients (ion-exchangers and plasticizer) to the bathing solution which is related to the distribution equilibria and diffusion rates. Another explanation can be related to the penetration of water molecules in the bathing solution to the membrane and consequently slow solvation of lipophilic salts so they are slowly leached out and limit the electrode life.

The proposed sensors showed rapid response within 20 s for drug solutions ≥ 10^{-4} mol L^{-1} and 30 s for dilute solutions (10^{-4}-10^{-7} mol L^{-1}). The sensors were stored and conditioned in 10^{-4} mol L^{-1} corresponding drug solution and washed carefully with bidistilled water between measurements .

A

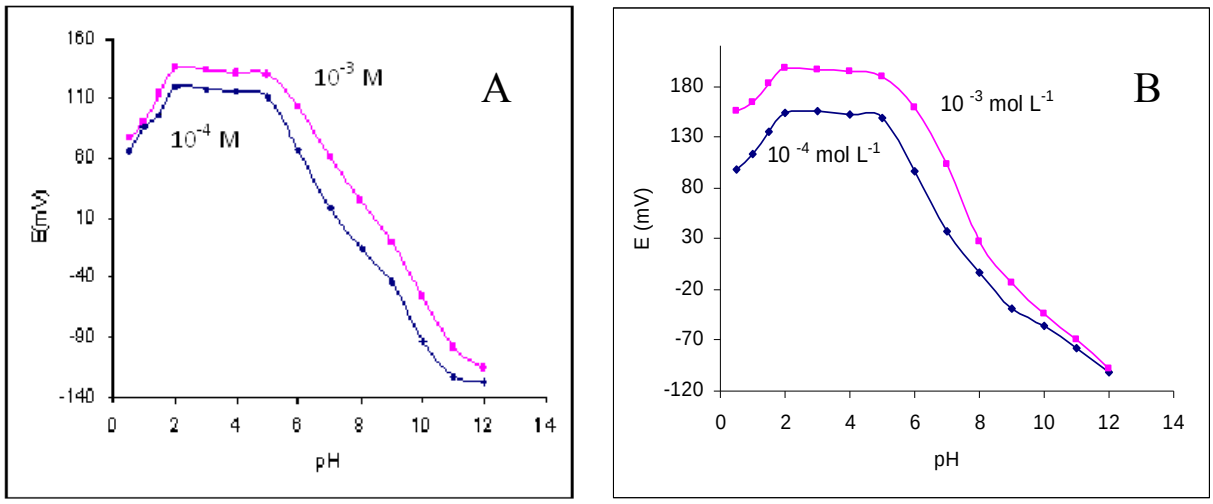


Fig. 2. Effect of pH on the potential responses of CLO-PVC (A) and PRX-PVC (B) membrane sensors

3.4. Potentiometric titration

Representative titration curves for the determination of the investigated drugs are shown in Fig. 3. It is noticed that as the concentration of the drug increases, the inflection of the curve at end point becomes sharper than low drug concentrations. So, these electrodes can be used successfully as indicator electrodes in potentiometric titrations of the investigated drugs. It also indicates that 1:1 [RK]: [Drug] are formed as seen from the curve.

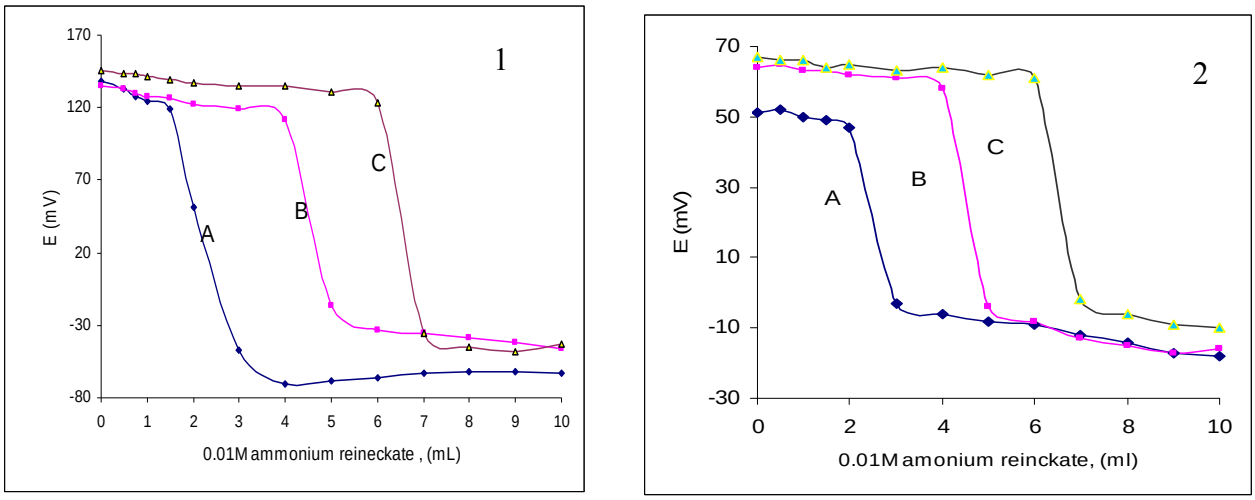


Fig. 3. Typical potentiometric titration of (A) 2.0 ml, (B) 4.0 ml and (C) 6.0 ml of 10^{-2} mol L^{-1} CLO and PRX drugs with 10^{-2} mol L^{-1} ammonium reineckate using CLO-RK (1) and PRX-RK (2) sensors

3.5. Selectivity of the electrodes

The selectivity of the ion-exchanger of membrane electrodes depends on the selectivity of the ion-exchange process at the membrane test solution interface and the mobility of the respective ions in the matrix of the membrane. The hydrophobic interactions between the primary ions and the PVC membrane are reflected by the values of the Gibb's energy of transfer for cations between the aqueous and membrane phases. The response of the electrodes towards different substances and ionic species was checked. The selectivity coefficients of the interfering cations were determined by the separate solution method which is a very simple method and provides a reasonable measure of the degree of the interference of foreign species, which may be present in the test solution [44].

From the value of the K_{ij} which is the measure of the electrode ability to demonstrate against interfering ions. From the results usually K_{ij} is smaller than (1) which means that ISEs are more selective to target ions.

Therefore, the sensors have been found to be chemically inert to other substances. The inorganic cations did not interfere owing to the differences in ionic size, and consequently their mobility and permeability, as compared with those of CLO and PRX drugs. The selectivity of the membrane to CLO and PRX drugs is due to the fact that insufficient interaction of the other substances with CLO and PRX-reinekate. The response of the sensor for different substances shows the best selectivity to CLO and PRX drugs (Table 2).

Table 2. Potentiometric selectivity coefficients of (Ki,jpot) for the two proposed drugs-PVC electrodes based on BES as mediator (i), with interfering ion (j)

Interfering ion, M	CLO-RK $K_{i,j}^{pot}$	PRX-RK $K_{i,j}^{pot}$
Na(I)	5.20×10^{-3}	10.10×10^{-3}
K(I)	6.50×10^{-3}	3.30×10^{-3}
Ca(II)	7.50×10^{-3}	6.00×10^{-3}
Mn(II)	6.50×10^{-3}	7.60×10^{-3}
Ba(II)	8.90×10^{-3}	7.60×10^{-3}
Co(II)	4.52×10^{-3}	5.03×10^{-3}
Mg(II)	6.23×10^{-3}	4.96×10^{-3}
Cu(II)	3.89×10^{-3}	4.67×10^{-3}
Zn(II)	8.56×10^{-3}	6.75×10^{-3}
Urea	6.99×10^{-3}	7.05×10^{-3}
Starch	7.50×10^{-3}	6.90×10^{-3}
Lactose	3.20×10^{-3}	3.80×10^{-3}
Sucrose	7.20×10^{-3}	2.70×10^{-3}
Fructose	4.20×10^{-3}	4.60×10^{-3}
Clomipramine	-	33.80×10^{-3}
Paroxetine	4.80×10^{-3}	-

4 . APPLICATIONS

CLO and PRX in pharmaceutical preparation were analyzed and their concentrations were determined using the respective electrodes. The results are listed in Table 3. The t-test and F-values obtained at the 95% confidence level and degree of freedom did not exceed the theoretical tabulated values indicating that there is no significant difference between accuracy and precision of the proposed and the official methods. Correlation coefficient values of 0.9963 and 0.9997, recovery from 97.25 to 101 and 99.75 to 100.6%, standard deviation

0.042-0.077 and 0.035-0.067 and variation coefficient 0.0449- 0.179 and 0.179-0.603 were obtained for CLO and PRX drug, respectively. The results outline that the proposed electrodes can be used to determine CLO and PRX drugs in pure samples or in pharmaceutical preparations with high accuracy and high percentage recovery without pretreatment procedures of the samples to minimize interfering matrix effect.

Table 3. Determination of chlomepramine and paroxetine in pharmaceutical prepararions using CLO-RK and PRX-RK sensors

Drug	[Drug] μg mL^{-1} Taken	[Drug] (n = 3) μg mL^{-1}						t-test	F-test
		Proposed method			Official method				
		Found	% Recovery	SD*	Found	% Recovery	SD*		
CLO-RK sensor: Anapramine	4.00 6.00 8.00	3.89 6.06 7.90	97.25 101.0 98.75	0.077 0.042 0.070	3.92 5.80 7.80	98.00 96.70 97.50	0.056 0.141 0.141	0.55 8.67 2.00	1.89 0.09 0.25
PRX-RK sensor: Xanadol	4.00 6.00 8.00	3.99 5.99 8.05	97.75 99.83 100.6	0.067 0.047 0.035	4.04 6.02 8.01	101.0 100.3 100.1	0.028 0.014 0.070	1.01 1.06 1.60	0.06 0.25 1.25

-Tabulated t–values at 95% confidence limit = 3.182 at degrees of freedom =3

- Tabulated F–values at 95% confidence limit = 9.12

The standard addition method was successfully applied for the analyses of the CLO and TRX drugs in tablets by the developed sensors (Table 4). Good correlation and recovery percentage between the results obtained and the claimed values were achieved. The accuracy of the proposed sensors was estimated by the recovery studies of CLO and PRX added to its tablet. The F- and t-tests at 95% confidence levels did not exceed the tabulated (theoretical) ones and no significant differences observed between the developed sensors and the method reported [45] with respect to precision and accuracy (Table 4). Thus, statistical analysis revealed that the proposed sensors are good and comparable with the reported methods.

4.1. Statistical evaluation of the proposed potentiometric method

The precision of the proposed PVC sensors, measured as percentage relative standard deviation (RSD%) was done. It is carried out by repeating the proposed method for determination of CLO and PRX drugs in their pharmaceutical preparations and the results obtained are listed in Table 4. The RSD% values for determination of CLO and PRX in Anapramine and Xanadol tables using CLO-RK and PRX-RK plastic membrane electrodes were found to be less than 2% indicating good precision of the proposed potentiometric method The accuracy of the proposed CLO-RK and PRX-RK sensors was calculated by the determination of CLO and PRX drugs in spiked Anapramine and Paroxitineplacebo samples prepared from serial concentrations of CLO and PRX reference standards. The results show the success of the proposed CLO-RK and PRX-RK sensors in the determination of CLO and PRX drugs in their pharmaceutical preparations without interferences from the coformulated adjuvants as indicated by the percentage recovery values (Table 4).

Table 4. Comparative analytical results of the proposed and official methods for the determination of CLO and TRX drugs in some pharmaceutical preparations

Sample	**Statistical parameter**	CLO-RK			PRX-RK		
		Direct potentiometry		Official method	Direct potentiometry		Official method
		Standard	Calibration addition method		Standard	Calibration addition method	
Anapramine (mg/tablet)	Mean%*	98.96	99.46	99.28	99.13	98.77	99.47
	N	0.443	0.672	0.637	0.532	0.879	0.728
	Variance	0.839	0.696	0.785	0.598	0.702	0.924
	SD	0.256	0.345	0.412	0.542	0.239	0.609
	SE	0.842	0.702	0.788	0.601	0.708	0.932
	RSD	1.33	0.452		0.233	0.705	
	T#	1.143	0.786		0.419	0.577	
	F##						
Xanadol (mg/tablet)	Mean%	99.05	98.69	99.73	99.68	99.09	99.58
	Variance	0.567	0.607	0.752	0.365	0.567	0.487
	SD	0.607	0.780	0.815	0.694	0.477	0.615
	SE	0.463	0.623	0.405	0.669	0.562	0.387
	RSD	0.612	0.786	0.817	0.699	0.480	0.618
	t#	0.731	6.644		0.283	0.794	
	F##	0.555	0.916		1.27	0.602	

*N = 5. # Tabulated t–values at 95% confidence limit = 3.182 at degrees of freedom = 5

Tabulated F–values at 95% confidence limit = 9.12 at degrees of freedom = 5

The linearity of CLO-RK and PRX-RK sensors is tested by plotting the relation between the electrode potential/mV and the logarithm of corresponding concentration of the drug under the previous optimized experimental conditions. The regression data, correlation coefficients (r) and other statistical parameter are listed in Table 1. Also the t- and F-values at 95% confidence limit are close to each other (tabulated values not exceed the theoretical one) indicating no significant difference between the proposed sensors and the official method with respect to precision and accuracy (Table 4) [45].

5. CONCLUSION

CLO and PRX-selective PVC membrane electrodes were described based on the ion-pair compound of drug-DBS or DOP. Their linear range, slopes and limit of detection indicates the sensitivity of the electrodes (Table 1). The effect of pH on the potential response indicated that larger influence of pH occurred when pH of the solution was in the range of 2~5.The method is simple, rapid selective and does not require expensive equipments. The proposed electrodes were successfully applied for the determination of clomipramine and paroxetine hydrochloride in pharmaceutical preparation.

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