Int. J. Electrochem. Sci., 6 **(2011)** 3036 - 3056

International Journal of ELECTROCHEMICAL SCIENCE
www.electrochemsci.org

Potentiometric Assay of Antipsychotic Drug (Ziprasidone Hydrochloride) in Pharmaceuticals, Serum and Urine

Vinod K. Gupta[1, 2 *], *Shilpi Agarwal*[1], *Barkha Singhal*[3]

[1]Department of Chemistry, Indian Institute of Technology Roorkee, Roorkee (U.K.) 247667 India
[2]Chemistry Department, King Fahd University of Petroleum and Minerals, Dhahran, Saudi Arabia
[3]School of Biotechnology, Gautam Buddha University, Greater Noida, (U.P.) 201308-India
[*]E-mail: vinodfcy@gmail.com;vinodfcy@iitr.ernet.in

Received: 14 April 2011 / *Accepted:* 6 June 2011 / *Published:* 1 July 2011

The membranes of ziprasidone hydrochloride-tetraphenyl borate, (Zp-TPB), chlorophenyl borate (Zp-ClPB), phosphotungstate (Zp_3-PT), ion associations as molecular recognition reagent dispersed in PVC matrix with dibutylphthalate as plasticizer have been proposed for quantification of ziprasidone hydrochloride. The performance characteristics revealed a fast, stable and liner response for ziprasidone over the concentration ranges of 8.5×10^{-6}-1.0×10^{-2} M, 3.9×10^{-6}-1.0×10^{-2}M, 7.7×10^{-7}-1.0×10^{-2} M ZpCl with cationic slopes of 57.0, 56.0, 58.5 mV/decade respectively. The solubility product of the ion-pair and the formation constant of the precipitation reaction leading to the ion-pair formation were determined conductometrically. The potentiometric determination of ziprasidone hydrochloride ion in different pharmaceutical preparations and biological fluids has been achieved without any interference from various excipients and diluents commonly used in drug formulations. Validation of the method shows suitability of the proposed electrodes for use in the quality control assessment of ziprasidone hydrochloride. The proposed potentiometric methods offer the advantages of high–throughput determination, simplicity, accuracy, automation feasibility and applicability to turbid and colored sample solutions.

Keywords: Membrane selective electrodes, pharmaceutical analysis, ion-pair, solubility product, neutral carriers.

1. INTRODUCTION

In the last few years there has been an overall increase in the use of antipsychotic drugs worldwide mainly of those considered as atypical. Atypical antipsychotic drugs are regarded as being safer and sometimes more effective than typical drugs, although cost is an important issue. Ziprasidone hydrochloride is described chemically as 5-[2-[4-(1,2-benzisothiazol-3-yl)-1-piperazinyl]ethyl]-6-

chloro-1,3-dihydro-2H-indol-2-one [1,2]. It is a novel atypical anti psychotic exhibits a potent highly selective antagonistic activity on the dopamine D_2 and serotonin ($5HT_2$) receptors. It exhibit high *in vitro* binding affinity for the dopamine D_2 and D_3, the serotonin $5HT_{2A}$, $5HT_{2C}$, $5HT_{1A}$, $5HT_{1D}$ and α1-adrenergic receptors and moderate affinity for the histamine H_1 receptor. The blockade of D_2 receptors in the mesolimbic area of the brain is thought to reduce hallucinations and delusions. Extrapyramidal symptoms (EPS) and cognitive deficits are thought to occur from dopamine blockade in the nigrostriatal and mesocortical pathways, respectively. The mechanism of action of ziprasidone as with other antipsychotic drugs effective in schizophrenia treatment, is still unknown. This drug is effectively used to treat the positive, negative and depressive symptoms associated with schizophrenia. Positive symptoms include visual and auditory hallucinations and delusions. Negative symptoms, which are harder to treat, include blunted affect, social withdrawal and lack of motivation. A literature survey reveals that the number of the analytical methods referring to the drug is relatively limited. The analytical methods that have been employed in the assessment of ziprasidone in pharmaceutical preparations and in biological fluids by using different techniques like spectrophotometry [3,4], HPLC [5-9], LC-MS [10,11]. The use of these techniques usually requires several time consuming manipulation steps, expensive instruments and professional training. Much effort is required in developing a procedure for the measurement of any drug substance is high sensitivity coupled with applications to biological fluids.

Because of ever increasing need for analytical methods with high sample throughput, low limits of detection and low maintenance cost, new methodologies are constantly being developed. Therefore, it is very imperative to develop a simple and suitable analytical method for the measurement of these drugs in bulk and pharmaceutical preparations. Developments in pharmaceutical analysis with ion-selective electrodes [12] have enabled the direct and selective measurement of the activity of various organic cations or anions of pharmaceutical interest, in most instances without prior separation of the active substance from the formulation matrix. These sensors offers the advantage of simple design and operation, reasonable selectivity, fast response, applicability to colored and turbid solutions and possible interfacing with automated and computerized systems. Recently, our research group has designed new ISEs for different pharmaceutically active substances with good analytical credentials [13-17]. No ion selective electrode has yet been used, as far as the authors are aware, for its determination. This led us to study its electroanalytical behavior in an attempt to develop a simple, sensitive rapid and reliable method for its determination in dosage form and biological fluids and the results were promising.

The present communication reports the results of preparation, characterization and application of ziprasidone selective electrodes based on incorporation of ziprasidone-tetraphenylborate, ziprasidone-chlorophenyl borate and ziprasidone-phosphotungstate (Fig. 1) as ion exchangers in poly (vinyl chloride) plasticized with dibutyl phthalate. This work describes an analytical method with acceptable analytical characteristics of suitability, reliability and feasibility. The electrodes have been used for the determination of the active ingredients in their respective pharmaceutical formulations without any prior separation and biological fluids such as plasma, urine samples with good reproducibility. The effects of membranes compositions, response time, selectivity and other factors have been investigated on electrodes performance are described.

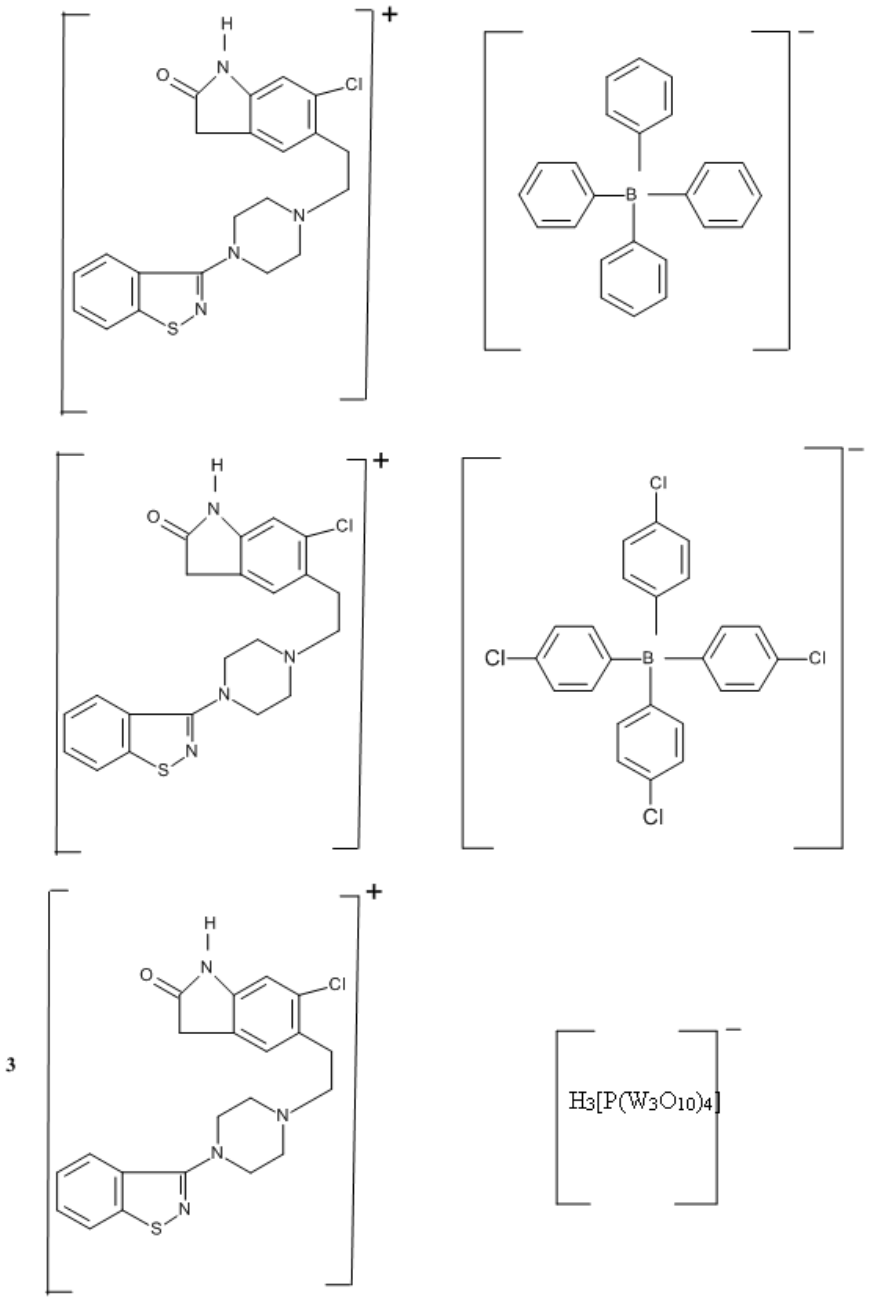

Figure 1. Structural formulae of ion association complexes of ziprasidone hydrochloride with PT, NaTPB and KTpClPB.

2. EXPERIMENTAL

2.1 Reagents and materials

The highest-grade commercially available reagents were used for the preparation of electrodes and double distilled deionized water was used throughout. Membrane components phosphotungstic

acid (PTA), sodium tetraphenyl borate (NaTPB), Potassium tetrakis (4-chlorophenyl) borate (KTpClPB), dioctylpthalate (DOP), *o*-nitrophenyloctyl ether (*o*-NPOE), dibutylpthalate (DBP) was obtained from Fluka. High molecular weight poly (vinylchloride) (PVC), used as the electrode membrane material and as a solvent for the membrane components, freshly distilled tetrahydrofuran (THF) was obtained from Merck.

The nitrate and chloride salts of all the inorganic cations used were of analytical grade and used without any further purification. The pharmaceutical preparations containing ziprasidone hydrochloride (Azona 20mg/tablet (Torrent pharma), Zipsydon 20 mg/tablet (Sun pharma) were purchased from local drug stores.

2.2 Preparation of ion-association complexes

The ion-association complexes Zp_3-PT, Zp-TPB, Zp-ClPB, were prepared by mixing stoichiometric amounts of 100 ml of 10^{-2} M (ZpCl) solution to the appropriate volume of 10^{-2} M solution each of PT, TPB, ClPB. The precipitates were filtered and washed thoroughly with deionized water for several times. Then precipitates were dried at room temperature for at least 48 h. and ground to fine powders. Elemental analyses were carried out to study the formation of these complexes.

2.3 Electrode Fabrication

The electrodes were fabricated as previously described by Thomas *et al* [18]. Membrane composition was studied by varying the percentage (w/w) of the ion exchanger, PVC and plasticizer until optimum composition that exhibits the best performance characteristics is achieved. Membrane cocktails were formulated by dissolving the required amount of ion -exchanger, PVC and various plasticizers in about 5 ml of THF.

The homogenous mixture was obtained after complete dissolution of all the components, concentrated by evaporating THF and it has poured into polyacrylate rings placed on a smooth glass plate. The viscosity of the solution and solvent evaporation was carefully controlled to obtain membranes with reproducible characteristics and uniform morphology and thickness otherwise have shown a significant variation. The membranes of 0.4-mm. thickness have mounted in glass bodies by careful removal from the glass plate. The electrode bodies were filled with 1.0×10^{-3} M ZpCl solution. The electrodes should be washed with deionized water before measurements. The potential measurements were carried out at 25 ± 1^{o}C using saturated calomel electrodes (SCE) as reference electrodes. Before use, all the ISE were impregnated for 24 hours in a drug solution with a concentration of 0.01 M. The potentials have been measured by varying the concentration of drug solution in the range 1.0×10^{-7}–1.0×10^{-2} M. Initial solutions were prepared by dissolving accurately weighed portions of drugs in twice distilled water and then diluting them to lower concentrations. The representative electrochemical cell for the electromotive force measurement was as follows:

Ag/AgCl | KCl (satd.) | 0.01 M ZpCl ‖ PVC membrane‖ test solution | Hg/Hg_2Cl_2 | KCl (satd.)

3. GENERAL PROCEDURES

3.1 Conductimetric determination of ZpCl

A volume containing 9.25-118.80 mg of ZpCl was transferred to a 50.0 ml volumetric flask and made up to the mark with double distilled water and the conductivity cell was immersed. Then 10^{-2} M PTA, NaTPB and KTpClPB were added and the conductance was measured subsequent to each addition of the reagent solution after thorough stirring. The conductance reading after each addition was corrected for dilution [19] by means of the following equation, assuming that conductivity is a linear function of dilution:

$$\Omega_{corr} = \Omega_{obs}[(v_1 + v_2)/v_1] \quad (1)$$

Where Ω is electrolytic conductivity, v_1 is the initial volume and v_2 is the volume of the added reagent (corr. = corrected and obs. = observed). A graph of corrected conductivity versus volume of the added titrant was constructed and the endpoint was determined.

3.2. Conductimetric determination of the solubility product of the ion associates

A series of solutions of different concentrations (c) was prepared for ZpCl, PT, NaTPB and KTpClPB. The measured conductivities of these solutions at 25°C were used to calculate the specific conductivities corrected for the effect of solvent and dilution, then the equivalent conductivities (λ) of the solutions were obtained. Straight-line plots of λ versus $\sqrt{c}$ were constructed and λ_0 for ZpCl, PT, NaTPB and KTpClPB were determined from the intercept of the respective line with the λ axis. The activity coefficients of the ions employed were taken as unity because all the solutions were sufficiently dilute (1.0×10^{-5} -1.0×10^{-2} M). The values of $\lambda_{0Zp\text{ -PT}}$, $\lambda_{0Zp\text{-TPB}}$, $\lambda_{0Zp\text{-ClPB}}$ were calculated using Kohlrausch's law of independent migration of ions [20].

The solubility (S) and solubility product (K_{sp}) of a particular ion associate were calculated using the following equations:

$$S = K_s \times 10000/\lambda_0 \text{ (ion associate)} \quad (2)$$

$$K_{sp} = S^2 \text{ for 1:1 ion associates} \quad (3)$$

$$K_{sp} = 27S^4 \text{ for 1:3 ion associates} \quad (4)$$

Where K_s, is the specific conductivity of the saturated solution of the ion associate determined at 25°C. The saturated solution was made by stirring a suspension of the solid precipitate in distilled water for 2 h at 25°C.

3.3. Measurement of selectivity:

The selectivity coefficients were determined by fixed interference method (FIM) [21], in which Nickolsky Eisenman equation was used:

(5)

$$K_{A,B}^{Pot} = \frac{a_A}{(a_B)^{z_A/z_B}}$$

Where, a_A is the activity of the primary ion A (Zp^+) at the lower detection limit in the presence of interfering ion B, a_B, the activity of interfering ion B and z_A and z_B are their respective charges.

The selectivity coefficients in case of species without charges were determined by matched potential method (MPM) [22]. In this method, the selectivity coefficient is defined as the activity ratio of primary and interfering ions that give the same potential change under identical conditions. At first, a known activity (a_{drug}) of the primary ion solution is added into a reference solution that contains a fixed activity of primary ions, and the corresponding potential change (ΔE) is recorded. Then, a solution of an interfering species is added to the reference solution until the same potential change is recorded. The change in potential produced at the constant background of the primary ion must be the same in both cases.

$$K_{Zp,J}^{pot} Z^+ = \frac{a_{drug}}{a_J} \quad (6)$$

Where, a_J is the activity of the interfering ion

3.4. Potentiometric determination of ZpCl:

ZpCl has been determined potentiometrically using the investigated electrodes by the standard addition method [23]. In standard addition method, known small increments of 1.0×10^{-2} M standard ZpCl solution were added to 50.0 ml aliquot samples of various concentrations (1.0×10^{-7}-1.0×10^{-2} M) of pure drug and pharmaceutical preparations. The change in potentials was recorded for each increment and used to calculate the concentration of ZpCl sample solution using the following equation:

$$C_x = C_s\left(\frac{Vs}{Vx+Vs}\right)\left(10^{n(\Delta E/S)} - \frac{Vx}{Vx+Vs}\right)^{-1} \quad (7)$$

Where C_x and V_x are the concentration and volume of the unknown, respectively, C_s, V_s are the concentration and volume of the standard, respectively, S is the slope of the calibration graph and ΔE is the change in potential due to the addition of the standards.

3.5. Ziprasidone assay in pharmaceutical preparations

Five tablets each of Azona, Zipsydon were accurately weighed and powdered in a mortar; the required amount from the tablet powder was dissolved in about 30 ml double distilled water and filtered in a 50 ml measuring flask. The residue was washed repeatedly and the volume was completed to the mark with same solvent. The contents of the measuring flask were transferred into a 100 ml beaker and subjected to potentiometric determination of ZpCl.

3.6. Ziprasidone assay in spiked urine samples

Blank urine samples were obtained from healthy male volunteers and different amounts of ZpCl and 5ml of urine were transferred to 50 ml measuring flask and completed to the mark by bidistilled water. The contents of the measuring flask were transferred to a 100 ml beaker and subjected to potentiometric determination of ZpCl by the standard addition method.

3.7. Ziprasidone assay in spiked serum samples

Human serum samples were obtained from healthy individuals and were stored frozen until the assay. An aliquot of the standard stock solutions of ZpCl was fortified with the human serum sample and this solution was diluted to 1.0 ml volume with acetonitrile in a 2 ml centrifuge tube. The tubes were vortexed for 10 min and then centrifuged for 3 min. The clear supernatant layer was a protein-free spiked human serum sample. The supernatant was taken carefully and appropriate volume of supernatant liquor was subjected to potentiometric determination by the standard addition method.

3.8. Potentiometric titration of ZpCl:

An aliquot of ZpCl (3.0×10^{-3} M- 7.5×10^{-3} M) was transferred into a 100-ml beaker and the solution was diluted to 50 ml with double distilled water and then titrated against a standard solution of PTA, NaTPB and KTpClPB using the investigated electrodes as indicator electrodes. The same method was applied for determination of ZpCl in the pharmaceutical preparations.

4. RESULTS AND DISCUSSION

4.1. Optimization of the ISE response

In the plastic membrane of an ion-selective electrode, the amount of lipophilic salt should be sufficient to obtain reasonable ionic exchange at the gel layer/test solution interface, which is responsible for the membrane potential. Besides the critical role of nature and amount of ion-\exchanger in preparing membrane selective electrodes some other important features of the PVC

membrane such as the nature of plasticizer, the plasticizer and PVC ratio and used are known to significantly influence the sensitivity and selectivity of ion- selective electrodes.

4.1.1. Evaluation of plasticizers performance

The introduction of high molecular PVC, as regular support matrix and traps for the sensed ions, creates a need for a plasticizer [24]. In the present investigation, DBP and DOP were chosen from diesters of dicarboxylic acids and *o*-NPOE was chosen as an example of the nitro aromatic group.

The selectivity of an ISE is codetermined by the solvation of the complexes between ionophore and primary and interfering ions, by desolvation of free ionophore in case of different stoichiometries of the complexes of primary and interfering ions, and by the solvation of interfering ions that do not form a complex with ionophore. Therefore, the plasticizers have a large influence on the selectivity of ion-selective electrodes [25]. Thus, we studied the effects of plasticizers on the selectivity of proposed electrodes plasticized with DBP, DOP, *o*-NPOE and the results are summarized in Table 1.

Table 1. Influence of plasticizers on selectivity pattern of the proposed electrodes

Cationic species	DOP			DBP			*o*- NPOE		
	Zp-ClPB	Zp_3-PT	Zp-TPB	Zp-ClPB	Zp-TPB	Zp_3-PT	Zp_3-PT	Zp-TPB	Zp-ClPB
Na^+	1.32	1.44	1.56	2.0	2.6	3.3	0.58	0.72	1.05
K^+	0.59	1.07	1.43	2.2	2.9	3.9	0.67	0.79	0.99
Mg^{2+}	1.2	1.39	1.50	2.4	3.5	4.0	0.89	1.06	1.12
Ca^{2+}	0.72	0.95	1.1	2.8	3.6	4.3	0.98	1.15	1.23
Lactose	0.26	0.44	0.68	2.7	3.8	4.5	1.05	1.09	1.18
Maltose	0.13	0.33	0.55	3.0	3.9	4.7	1.09	1.20	1.30
Alanine	0.77	0.88	0.99	3.2	4.0	4.8	1.18	1.32	1.37
Glycine	1.35	1.68	1.77	3.4	4.3	4.9	1.26	1.25	1.42
Sucrose	1.49	1.72	1.99	3.5	4.5	5.1	1.35	1.47	1.64
Urea	0.16	0.29	0.59	3.7	4.7	5.3	1.45	1.58	1.55

In the present investigation, DBP was chosen as plasticizer from diesters of carboxylic acids. With PVC, the diesters of carboxylic acids were found to be the optimum plasticizers; they plasticized the membrane, dissolve the ion association complex, and adjust both permittivity of the final organic membrane and mobility of the ion exchange sites. Such adjustments influence the partition coefficient of the studied drug with subsequent effect on electrode sensitivity. The interference by other species was markedly increased in plasticizer with high polarity, such as *o*-NPOE, thus low polarity environments provided more appropriate conditions for complexation.

Several compositions for the electrodes were investigated in which the ion-exchanger percentage ranged from 2.4 % to 6.9 % for Zp-TPB, Zp-ClPB and from 2.4 % to 9.0 % for Zp_3-PT

electrodes. For each composition, the electrodes were repeatedly prepared four times. The preparation process was highly reproducible as revealed by the low relative standard deviation values of the slopes obtained employing the prepared membranes. The best performances were obtained using compositions of 3.60 % Zp-TPB, Zp-ClPB, 48.2% PVC and 48.2 % DBP for Zp_3-PT, 9.08 % Zp_3-PT, 45.5 % PVC and 45.5 % DBP. The above optimum compositions were used to prepare membrane electrodes for all further investigations.

4.1.3. Response characteristics of proposed electrodes

Electrochemical performance characteristics of the proposed electrodes were systematically evaluated according to IUPAC standards [26, 27]. The response characteristics of the three electrodes are summarized in Table 2.

Table 2. Response characteristics of Ziprasidone based electrodes

Parameter	Zp_3-PT	Zp-TPB	Zp-ClPB
Composition (%) (Ion-associate-PVC- DBP)	(9.0 – 45.5- 45.5)	(3.6 – 48.2 - 48.2)	(3.6 – 48.2 - 48.2)
Slope (mV/ decade)	58.5	57.0	56.0
Linearity range (M)	7.7×10^{-7}-1.0×10^{-2}	8.5×10^{-6}-1.0×10^{-2}	3.9×10^{-6}-1.0×10^{-2}
Detection limit (M)	4.6×10^{-7}	5.2×10^{-6}	2.7×10^{-6}
Response time (s)	15	25	20
Life span (days)	35-40	15-20	15-20
Working pH range	3.0-7.0	3.0-8.0	3.0-7.5

4.1.4. Effect of internal solution

It is well known that the internal reference solution affects substantially the characteristics of the electrodes. There was negligible response with high KCl concentrations. The slope of the response was close to the theoretical value with equimolar solutions of drugs with KCl as the inner solution, but the linear range was very narrow. The best results in terms of characteristics of the electrodes were obtained with an inner solution containing only 1.0×10^{-2} M of drugs solution. The former results might be due to the fact that the Donnan equilibrium was reached at membrane/inner solution interface and an electrical potential was generated (Donnan potential) necessary to develop the membrane potential. Thus it was probably not the case when the inner solution contained KCl 0.01 M or 0.1 M.

4.2. Influence of soaking and Lifetime

It has been observed that high solubility of some drug complexes in aqueous media and leaching of these complexes from the solvent polymeric membrane phase significantly affect the life time and the potentiometric response characteristics of sensors. A complicated approach to overcome the leaching problems was a repeated soaking of the membranes in the test solution [28, 29]. The freshly prepared electrodes must be soaked to activate the surface of the membrane to form an infinitesimal thin gel layer at which ion exchange occurs. This preconditioning process requires

different time depending on diffusion and equilibration at the electrode test solution interface fast establishment of equilibrium is certainly a sufficient condition for fast potential response [30].

For the present electrodes, the presoak times were 24 h with slopes of 58.5, 57.0 and 56.0 mV/ decade for Zp_3-PT, Zp-TPB and Zp-ClPB, respectively.

Nevertheless, continuous soaking of the electrodes in 10^{-2} M ZpCl solution affects negatively their response to the Zp^+. This is attributed to leaching of the active ingredients (ion-exchangers and plasticizer) to the bathing solution. It was notice2d that the slopes of the calibration graphs obtained by the preconditioned electrodes were nearly constant for 15-20 days in case of Zp-TPB, Zp-ClPB electrodes and 35-40 days in the case of Zp_3-PT, then the slopes of the three electrodes started to decrease gradually reaching 50.8, 47.5, 43.7 mV/ decade for Zp_3-PT, Zp-TPB, Zp-ClPB, respectively. However, it was noted that, in all cases, electrodes which have been kept dry in a closed vessel and stored in a refrigerator showed a good preservation of the slope values and response properties extending to several months.

The decrease in the efficiency of the electrode is due to a diminished Zp^+ ion exchange rate on the membrane gel layer-test solution interface, which is responsible for the membrane potential.

4.3. Regeneration of the electrodes

The above discussion reveals that soaking of the electrodes in the drug solution for a long time has a negative effect on the response of the membranes towards its ion. The same effect appears after working with the electrode for a long time.

The regeneration of the electrodes was tried simply by reformation of the ion exchangers on the external gel layer of the membrane and this was successfully achieved by soaking the exhausted electrodes for 20 h in a solution of 1.0×10^{-2} M in PTA, TPB, ClPB followed by soaking for 14 h in 1.0×10^{-2} M ZpCl solution. The slopes of the exhausted electrodes were found to be 51.0, 47.7, 43.0 mV/decade but after regeneration they reached 54.2, 51.3. 52.9 mV/decade for Zp_3-PT, Zp-TPB, Zp-ClPB electrodes, respectively.

4.4 Potential-pH profile of the sensor

The effect of pH of the test solution (1.0×10^{-2} - 1.0×10^{-4} M ZpCl) on Zp_3-PT Zp-TPB, Zp-ClPB electrode potentials was investigated by following the variation in potential with change in pH by addition of very small volumes of HCl and NaOH (each 0.1- 1.0 M). The results indicated that the electrode displayed no response to the pH change in the range 3.0-7.0, 3.0-8.0, 3.0-7.5 for Zp_3-PT, Zp-TPB, Zp-ClPB respectively (Fig. 2). However, outside this range the electrode responses at pH < 3.0 seems ascribable to penetration of chloride ion in the membrane gel layer or the formation of diprotonated species. The decrease occurring at higher pH values is most probably attributed to the formation of the free Ziprasidone base in the solution, leading to a decrease in the concentration of ziprasidone cation.

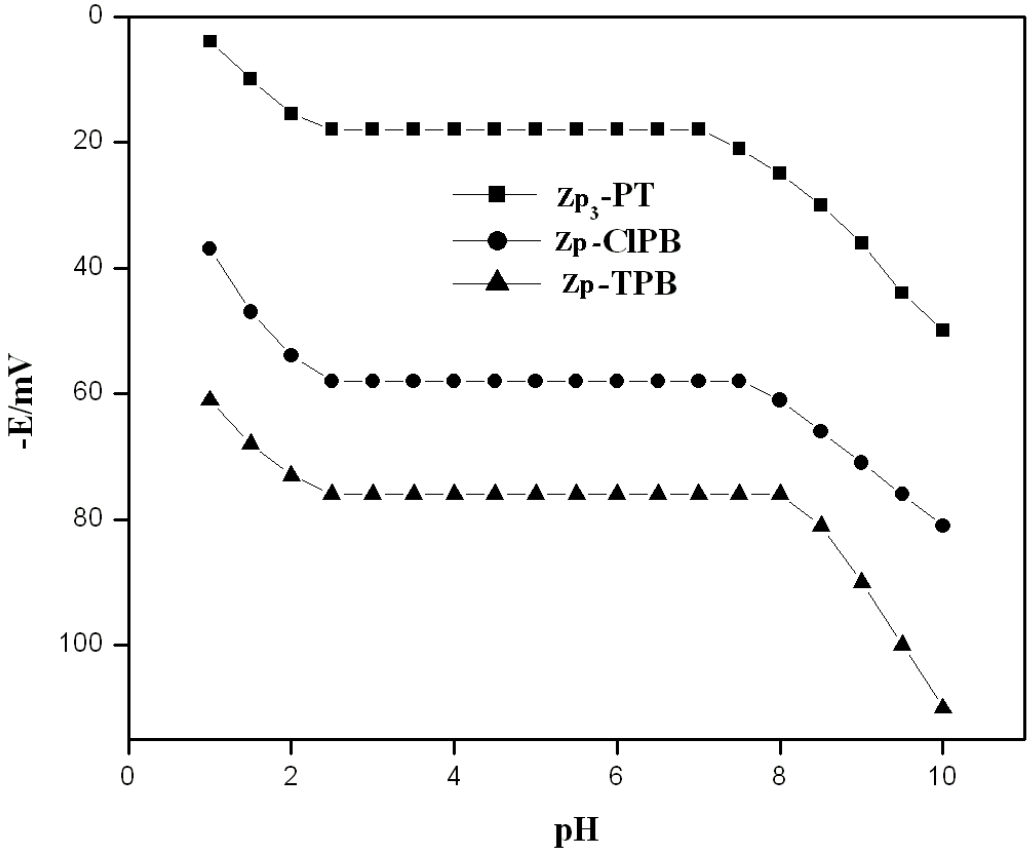

Figure 2. Potential–pH profile of ziprasidone based electrodes.

4.5. The Dynamic response time behavior of the proposed electrodes

It is well known that the dynamic response time of a sensor is one of the most important factors in its evaluation. To measure the dynamic response time of the proposed electrodes the concentration of the test solution has been successively changed from 1.0×10^{-4} M to 1.0×10^{-2} M. The required time for the electrode to reach value within ± 1 mV from the final equilibrium potential after increasing the level of the drug concentration to 10-fold be fairly short and 90 % of the final steady potential was reached after up to 15 s for Zp_3- PT, 20 s for Zp-ClPB and 25 s for Zp-TPB electrodes.

4.6 Conductimetric studies of pure solution of drug

Conductance measurements are used successfully in quantitative conductimetric titration of systems in which the conductance of the solution varies before and after the equivalence point. The system under investigation showed a regular rise in conductance up to the equivalence point where a sudden change in the slope occurs. The results of the drug determination presented in Table 3 showed that good recoveries and low standard deviations were obtained. The optimum concentration ranges for Zp^+ determination are 9.25–97.0, 10.5–96.5 and 23.1–118.8 mg with mean recovery values of 99.00–

99.69, 98.09–99.74 and 98.41–99.25 % with coefficients of variation of 0.19-0.56, 0.17-0.75, and 0.24-0.82 for Zp-TPB, Zp-ClPB and Zp_3-PT electrodes respectively at which sharp inflections and stable conductance readings were obtained.

Table 3. Conductimetric determination of Ziprasidone hydrochloride

		Ziprasidone hydrochloride		
Additive	Taken (mg)	Found (mg)	Recovery (%)	RSD (%)
ClPB				
	10.50	10.30	98.09	0.75
	25.00	24.80	99.20	0.32
	50.70	50.30	99.21	0.54
	70.80	70.40	99.43	0.25
	96.50	96.25	99.74	0.17
PT				
	23.10	22.90	99.13	0.51
	53.60	53.10	99.00	0.24
	80.50	79.40	99.25	0.35
	105.50	104.20	98.76	0.66
	118.80	117.0	98.41	0.82
TPB				
	9.25	9.20	99.45	0.56
	22.00	21.80	99.00	0.23
	48.50	48.20	99.38	0.45
	72.50	72.10	99.44	0.38
	97.00	96.70	99.69	0.19

4.7. Solubility products of ion associates

The determination of the solubility product of a precipitate is important [31] since its reciprocal is approximately equal to the equilibrium constant of the precipitation reaction leading to the ion-pair formation. It is noteworthy to mention that the solubility of an ion-exchanger is one of the main factors controlling the life span of the sensor which incorporate this ion-exchanger as electro active material. The solubility products of the ion associates were found to be 1.25×10^{-8}, 4.55×10^{-11}, and 1.88×10^{-6}, for Zp_3-PT, Zp-ClPB, and Zp-TPB electrodes, respectively. Consequently, the equilibrium constants of the ion-associate formation reaction can be calculated as follows:

$$Zp + ClPB \qquad k = 8.5\times 10^{7}$$

$$3\ Zp + PT \qquad k = 6.3\times 10^{9}$$

$$Zp + TPB \qquad k = 1.6 \times 10^{6}$$

These equilibrium constant values are very high, indicating that the degree of completeness of the ion-associate formation reaction is 99.9 %. In the equilibrium, the solubility product of the undissociated ion associate in water (i.e. the intrinsic solubility) was omitted as this term makes a negligible contribution to the total solubility because the ion associates are sparingly soluble in water and its saturated solution is, therefore, very dilute [32, 33].

4.8 Selectivity of the electrodes

The mechanism of ISE is based on the fact that the ion pair or ion associate complex is formed between drug and ion exchanger; it is dissolved in the organic phase (membrane) and then placed between the two aqueous phases, the sample solution containing both analytes (A^+) and interfering ion (B^+) and internal reference solution. An ion exchange reaction takes place in the membrane surface according to

$$A^+_m + B^+_s \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots A^+_s + B^+_m$$

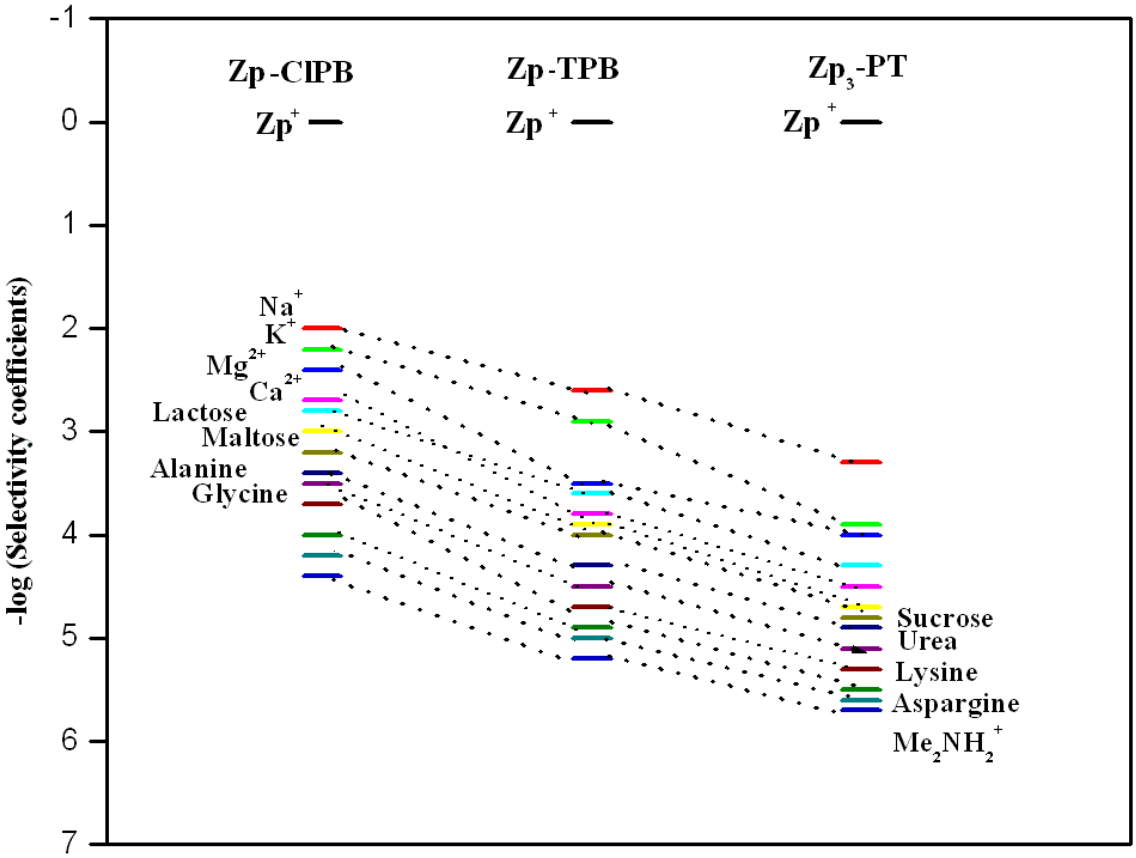

Figure 3. Selectivity of ziprasidone based electrodes.

Then, a constant concentration of these species is soon established in the solution, rather than in membrane, since diffusion inside the membrane is much slower than that in solution. The selectivity of the membrane therefore depends on the selectivity coefficient for A^+ with respect to B^+. The influence of some organic cations, sugars, amino acids, vitamin and urea on the ZpCl was investigated (Fig. 3). The selectivity coefficients of the proposed electrodes reflect a very high selectivity of the investigated electrodes for the Zp^+.

The mechanism of selectivity is mainly based on the stereospecificity and electrostatic environment, and is dependent on how much matching is present between the locations of the lipophilic sites in the two competing species in the bathing solution side and those present in the receptor of the ion exchanger [34]. The inorganic cations do not interfere because of differences in ionic size, mobility and permeability due to the fact that the smaller the energy of hydration of the cation, the greater the response of the membrane.

5. ANALYTICAL APPLICATIONS

In order to assess the applicability of the proposed electrodes, the methods were applied for determination of ziprasidone in its pharmaceutical preparations and in different biological fluids such as serum and urine.

5.1. Determination of Ziprasidone in *pharmaceutical preparations*

The proposed procedure was successfully applied for the assay of ziprasidone in Azona, Zipsydon tablets.

The recoveries of ziprasidone tablets were compared with those obtained by a standard method which involves a voltammetric [3] and RP-HPLC procedures [4] (Table 4). No interference was found from the excipients in the tablets analyzed by the present method. This means that the proposed procedure should be applicable to the analysis of this and other similar type of formulation products that contain ziprasidone with great success.

5.2. Determination of Ziprasidone in spiked urine samples

Determination of analytes present in physiological matrices, such as urine or serum, without any preliminary treatment by electroanalytical methods is more interesting. The determination of ziprasidone in spiked human urine and serum was chosen as a practical example. The average recovery of ziprasidone in spiked human urine was found to be 99.5 % and 98.2 %, 98.1 %. This confirms the good selectivity of the method. Results obtained for the determination of ziprasidone in urine samples are presented in Table 5.

Table 4. Assay of Ziprasidone in pharmaceutical preparations

	Azona		**Zipsydon**	
	Standard addition	Potentiometric titration	Standard addition	**Potentiometric titration**
Taken (M)				
	1.2×10^{-3}	3.0×10^{-3}	1.2×10^{-3}	**3.0×10^{-3}**
	2.2×10^{-3}	5.5×10^{-3}	2.2×10^{-3}	**5.5×10^{-3}**
	4.8×10^{-3}	7.8×10^{-3}	4.8×10^{-3}	**7.8×10^{-3}**
	6.0×10^{-3}		6.0×10^{-3}	
Zp_3-PT				
Recovery (%)	98.7	97.1	97.0	**101.2**
	97.5	98.5	98.4	**99.5**
	99.6	99.1	98.9	**96.4**
	98.2		97.3	
R.S.D. [a] (%)	0.18	0.87	1.07	**0.37**
	1.01	0.65	0.55	**0.15**
	0.48	0.27	0.61	**1.19**
	0.75		0.98	
Zp-TPB				
Recovery (%)	99.8	99.4	99.9	**98.1**
	98.2	97.2	100.4	**96.3**
	97.1	100.2	97.9	**100.1**
	98.3		98.0	
R.S.D. [a] (%)	0.21	0.16	0.22	**0.44**
	0.46	0.94	0.65	**0.80**
	0.99	0.77	0.97	**0.26**
	0.35		0.39	
Zp-ClPB				
Recovery (%	98.7	99.3	100.5	**97.9**
	97.7	97.4	99.6	**99.0**
	96.0	98.9	97.5	**98.1**
	99.2		96.0	
R.S.D. [a] (%)	0.29	0.16	0.79	**0.46**
	0.76	0.58	0.17	**0.08**
	1.02	0.30	0.69	**0.22**
	0.10		**1.06**	

Table 5. Determination of Ziprasidone hydrochloride in spiked urine samples by standard addition method

Taken (M)	Zp_3-PT		Zp-TPB		Zp-ClPB	
	Recovery (%)	R.S.D. (%)	Recovery (%)	R.S.D (%)	Recovery (%)	**R.S.D. (%)**
2.0×10^{-4}	98.3	1.0	97.3	1.4	97.8	**1.3**
5.0×10^{-4}	98.7	0.76	99.1	0.95	98.5	**0.88**
8.0×10^{-4}	**101.6**	**0.93**	**98.4**	**1.02**	**98.1**	**0.96**

5.3. Determination of Ziprasidone in spiked serum samples

The proposed method was also successfully applied for the determination of ziprasidone in serum samples, and the results are shown in Table 6. No extraction or pretreatment steps, other than the centrifugal separation of protein, were required prior to the assay of the drugs. The percentage of recovery of ziprasidone was calculated to be 97.8 % and 98.7 % and 98.8 %. Good recoveries of ziprasidone are obtained in spiked serum samples by the present method.

Table 6. Determination of Ziprasidone hydrochloride in spiked serum samples by standard addition method

Taken (M)	Zp_3-PT		Zp-TPB		Zp-ClPB	
	Recovery (%)	R.S.D. (%)	Recovery (%)	R.S.D (%).	Recovery (%)	**R.S.D. (%)**
2.0×10^{-4}	97.6	1.12	98.0	0.96	100.3	**0.77**
5.0×10^{-4}	97.0	1.24	98.9	0.85	98.2	**0.89**
8.0×10^{-4}	**98.8**	**0.87**	**99.2**	**1.17**	**97.7**	**1.03**

5.4. Statistical evaluation of results

The results of the pharmaceutical preparations were compared with the published procedures (Table 7). The results are in good agreement with those obtained from the reference method. Student's t-test and F-test (at 95 % confidence level) were applied [35-126]. The results show that the calculated t- and F- values did not exceed the theoretical values. The results demonstrate the validity of the proposed method for the determination of ziprasidone in tablet dosage forms.

Table 7. Statistical comparison between results of pharmaceutical preparations on applying the proposed and reference methods

	Parameters					Reference methods	Parameters		
	Mean recovery	S.D.	R.S.D	F- ratio (9.28)[a]	t-Test (2.447)[b]	RP-HPLC[4]	Method	Mean recovery	**S.D.**
Zp_3-PT							Standard addition	99.68	**1.23**
Azona	98.5	1.18	1.21	0.43	0.27	Voltammetry[3]	Standard addition	99.27	**1.18**
Zipsydon	97.9	0.95	1.03	0.67	0.38				
Zp-TPB									
Azona	98.3	1.05	1.09	0.55	0.21				
Zipsydon	99.0	1.20	1.24	0.42	0.16				
Zp-ClPB									
Azona	97.8	1.25	1.29	0.89	0.55				
Zipsydon	98.4	1.07	1.13	1.21.	0.98				

5.5. Recovery of Ziprasidone in pharmaceutical preparations at different expiry dates

Among all investigated electrodes in the present communication, Zp_3-PT displayed higher selectivity and stability.

Table 8. Assay of Ziprasidone hydrochloride in different pharmaceutical preparation at different expiry date by applying standard addition method for Zp_3-PT electrode

Taken (mg)	Newly manufactured batch		One month before its expiration date		One month after its expiration date		Six month after its expiration date	
Azona	Recovery (%)	RSD (%)	Recovery (%)	RSD (%)	Recovery (%)	RSD (%)	Recovery (%)	RSD (%)
1.2×10^{-3}	97.5	0.63	98.7	0.73	101.5	1.38	88.5	1.59
2.2×10^{-3}	98.2	0.75	100.2	0.99	100.8	1.08	87.7	1.72
4.8×10^{-3}	99.6	0.79	101.3	1.12	100.6	1.19	90.2	1.18
6.0×10^{-3}	98.0	0.82	98.9	0.84	98.7	1.29	86.8	1.87
Zipsydon								
1.2×10^{-3}	98.7	0.70	98.6	0.67	99.2	1.40	91.5	1.21
2.2×10^{-3}	97.9	0.66	100.4	1.10	101.5	1.32	89.4	1.57
4.8×10^{-3}	99.4	0.94	100.6	1.08	101.7	1.26	88.0	1.66
6.0×10^{-3}	98.3	0.87	99.1	0.88	98.5	1.04	87.3	1.85

So the analytical applicability Zp_3-PT was further assessed for pharmaceutical preparations of four batches of different expiry dates (Table 8) show that the concentration of ziprasidone hydrochloride was not affected by time except after six months from the expiration date where the concentration begin to decrease and the recoveries were ranged from 88.5-90.2, 87.3-91.5, 86.5-90.7 % with relative standard deviation 1.18-1.87, 1.21-1.85, 1.38-1.75 for pharmaceutical preparations, respectively.

6. CONCLUSION

The present study shows that ISEs are very promising platforms and offer an attractive solution for investigation of ziprasidone hydrochloride over other sophisticated analytical methodologies. The proposed potentiometric membrane electrodes have good analytical credentials. The construction of the electrodes is simple, fast and reproducible and assures the reliable response characteristics. The proposed electrode was successfully applied in the assay of pharmaceutical formulations and biological fluids with high accuracy and percentage recovery. The proposed sensor open a new very interesting field in the application of ISEs in the assay of the drugs in true biological samples containing the metabolites of the drugs and their validation which is must for quality control of drugs in pharmaceutical industry. Thus, the proposed methods proved to have precision and accuracy adequate for the reliable analysis of ziprasidone.

ACKNOWLEDGEMENT
The authors acknowledge Indian Institute of Technology Roorkee, Roorkee for supporting this work.

References

1. Al Dirbashi OY, Aboul-Enein HY, Al Odaib A, Jacob M, Rashed MS Biomed *Chromatogr* 20(2005)365–368.
2. Sachse J, Hartter S, Hiemke C *Ther Drug Monit* 27(2005)158–162.
3. P.K. Choudhary, P.K. Sharma, A.K. Mathur, P. Ramnani and P. Jain, *Orient J. Chem.*, (2006) 21.
4. G. Srinubabu, B. Sudha Rani and J.V.L.N. Seshagiri Rao, *E. J. Chem.*, 3 (2006) 9-12.
5. N.L. Prasanthi and N. Rama Rao, *Int. J. Pharm. Pharm. Sci.*), 2 (2010)120-122.
6. G. Zhang, A.V. Terry and M.G. Bartlett*, J. Chromatogr.* B, 856(2007) 20-28.
7. B. Sudha Rani and P. Venkata Reddy, *E. J. Chem.*, 3(2006) 169-172.
8. R.F. Suckow, M. Fein, C.U. Correll and T.B. Cooper*, J. Chromatogr.* B, 799(2004) 201-208.
9. G. Zhang, A.V. Terry and M.G. Bartlett*, J. Chromatogr.* B, 858(2007) 276-281.
10. M. Aravagiri, S.R. Marder and B. Pollock, *J. Chromatogr.* B, 847(2007) 237-244.
11. H. Kirchherr and W.N. Kuhn-Velten, *J. Chromatogr.* B, 843(2006) 100-113.
12. Abdel-Ghani N T, Shoukry A F, El-Nashar R M, *Analyst* 126(2001)79-82.
13. Gupta, V.K., Singh, A.K., Gupta, B., *Combinatorial Chem. High throughput screening*, *10*(2007) 583-594.
14. V.K. Gupta, M.K. Pal, A.K. Singh, *Electrochim. Acta*, 55*(*2010) 1061-1066.
15. V.K.Gupta, A.K. Singh, B. Gupta, *Anal. Bioanal. Chem.*, *389*,(2007) 2019-2028.

16. V.K.Gupta, A.K. Singh, B. Gupta, *Combinatorial Chem. High throughput screening*, *10*, (2007)560-570.
17. V.K. Gupta, M.K. Pal, A.K. Singh., *Electrochim. Acta*, *54* (2009) 6700-6706.
18. L.L. Antropov; *Theoretical Electrochemistry*, *Mir,* Moscow, 1977.
19. F.J. Sa'ez de Viteri, D. Diamond, *Analyst,* 119(1994) 749.
20. Y. Umezawa, P. Buhalmann, K.Umezawa, K. Thoda, S. Amemiya, *Pure Appl. Chem., 72,*(2000) 1851.
21. E. Baumann, *Anal. Chim. Acta, 42*,(1986) 127.
22. E. Bakker, P. Buhlmann, E. Pretsch, *Chem. Rev., 97,* (1997)3083.
23. T. Sokalski, T. Zwickl, E. Bakker, E. Pretsch, *Anal. Chem., 71,*(1999) 1210.
24. IUPAC, *Analytical Chemistry Division, Commission on Analytical Nomenclature*, *Pure Appl. Chem., 8*,(1976)129.
25. IUPAC, *Analytical Chemistry Division, Pure Appl. Chem.,* 67(1995)507.
26. M.M. Zareh, *Mikrochim. Acta, 126*, (1997) 271.
27. A.F. Shoukry, Y.M. Issa, M.S. Rizk, R.M. El-Nashar, *Anal. Lett*., *29,* (1996)1463.
28. E. Linder, K. Toth, E. Pungor, Chemical *Rubber Company (CRC) Press, Boca Raton*, USA, 1988.
29. E. Pungor, K. Toth, *Analyst*, *95,*(1970) 625.
30. A.F. Shoukry, *Analyst, 113*(1988) 1305.
31. H.M. Irving, R.J.P. Williams, *Analyst,* 77(1952) 813.
32. R.D. Armstrong, G. Horvai, *Electrochim. Acta, 35*(1990) 1.
33. J.C. Miller, J.N. Miller, Statistics *for Analytical Chemistry, second ed., Ellis Horwood*, Chichester, 1991, 260.
34. V. K. Gupta and P. Kumar, *Anal. Chim. Acta*, 389(1999) 205-212
35. A.K. Singh, V. K. Gupta and Barkha Gupta, *Anal. Chim. Acta*, 585(1) (2007)171-178
36. V. K. Gupta, A. K. Singh and Barkha Gupta, *Anal. Chim. Acta*, 575(2)(2006) 198-204
37. V. K. Gupta and A. Rastogi .*J. Hazardous Materials* 152(1) (2008) 407-414
38. R. Prasad, V. K. Gupta and Azad Kumar, Anal. Chim. Acta , 508(1),(2004) 61-70
39. S. K. Srivastava, V. K. Gupta and S. Jain, *Electroanalysis*, 8 (1996) 938.
40. S. K. Srivastava, V. K. Gupta and S. Jain, *Anal. Chem*. 68(1996)1272.
41. V. K. Gupta, S. Jain and U. Khurana, *Electroanalysis*, 9 (1997) 478
42. V. K. Gupta, A. K. Jain, L. P. Singh and U. Khurana, *Anal. Chim. Acta*, 355(1997)33.
43. A.K. Jain, V. K.Gupta, U. Khurana and L. P. Singh, *Electroanalysis*, 9 (1997) 857.
44. A.K. Jain, V. K. Gupta, L. P. Singh and U. Khurana, *Analyst*, 122(1997) 583.
45. V. K. Gupta, S. Jain and U. Khurana, *Electroanalysis*, 9 (1997) 478.
46. A.K. Jain, V. K.Gupta and L. P. Singh, *Analytical Proceedings including Analytical Communications*, 32 (1995) 263.
47. A. K. Jain, V. K.Gupta, B. B. Sahoo and L. P. Singh, *Analytical Proceedings including Analytical Communications* ,32 (1995) 99.
48. S. K. Srivastava, V. K. Gupta and S. Jain, *Analyst*, 120 (1995) 495.
49. V. K. Gupta, M.M. Antonijevic, S.Chandra and S.Agarwal, *Sensors,* 2 (2002) 233.
50. V. K. Gupta, R. N. Goyal and R. A. Sharma, *Electrochim. Acta,* 54 (2009)4216.
51. V. K. Gupta, R. N. Goyal, A. K. Jain and R. A. Sharma, *Talanta,* 78(2009) 484.
52. R. N. Goyal, V. K. Gupta, Neeta Bachheti, R. A. Sharma, *Electroanalysis,*20(2008)757.
53. R. N. Goyal, V. K. Gupta and N. Bachheti, *Anal. Chim. Acta,* 597(2007) 82.
54. R. N. Goyal, V. K. Gupta, M. Oyama and N. Bachheti, *Talanta,* 72(2007) 976.
55. V.K.Gupta, D.K.Chauhan, V.K.Saini, S. Agarwal, M. Antonijevic and H. Lang, *Sensors*, 3(2003) 223.
56. R. N. Goyal, V. K. Gupta and S. Chatterjee, *Anal. Chim. Acta,* 657(2010)147.
57. R. N. Goyal, V. K. Gupta and S. Chatterjee, *Biosensors and Bioelectronics,* 24(2009)3562.

58. R. N. Goyal, V. K. Gupta and S. Chatterjee, *Biosensors and Bioelectronics,* 24(2009) 1649.
59. R. N. Goyal, M. Oyama, V. K. Gupta, S. P. Singh and S. Chatterjee, *Sens. Actuat.B,* 134(2008)816.
60. R. N. Goyal, V. K. Gupta and S. Chatterjee, *Talanta*, 76(2008)663.
61. R. N. Goyal, V. K. Gupta and S. Chatterjee, *Electrochim. Acta,* 53(2008) 5354.
62. V. K. Gupta, A. K. Jain, S. Agarwal and G. Maheshwari, *Talanta,* 71 (2007) 1964.
63. V. K. Gupta, R. N. Goyal, S. Agarwal, P. Kumar and N. Bachheti, *Talanta*, 71(2) (2007) 795.
64. V. K. Gupta, A. K. Jain and G. Maheshwari, *Chemia Analityczna-Chemical analysis*, 51(2006)889.
65. V. K. Gupta, A. K. Jain and P. Kumar, *Electrochim. Acta,* 52(2) (2006)736.
66. V. K. Gupta, A. K. Jain, G. Maheshwari and H. Lang*, Sens. Actuat. B,* 117(1) (2006) 99.
67. V. K. Gupta, R. N. Goyal, M. A. Khayat, P. Kumar and N. Bachheti, *Talanta,* 69(5) (2006) 1149.
68. A.K. Jain, V.K. Gupta and J.R. Raisoni, *Talanta,* 69(4) (2006)1007.
69. V. K. Gupta, A. K. Singh, S. Mehtab and B. Gupta, *Anal. Chim. Acta,* 566(1) (2006) 5.
70. V. K. Gupta, S. Agarwal, A. Jakob and H. Lang, *Sens. Actuat. B,* 114(2) (2006) 812.
71. R. N. Goyal, V.K.Gupta, A. Sangal and N. Bachheti, *Electrochemistry Communications,* 8(2006) 65.
72. R. N. Goyal, V. K. Gupta, A. Sangal and N. Bachheti, *Electroanalysis,* 17 (24) (2005) 2217.
73. V. K. Gupta, S. Chandra and H. Lang, *Talanta,* 66(3) (2005) 575.
74. V. K. Gupta, R. Ludwig, S. Agarwal, *Anal. Chim. Acta,* 538(1-2) (2005) 213-218.
75. R. K. Bera, S.K Sahoo, S. K. Mittal and A. Kumar, *Int. J. Electrochem. Sci.,* 5(2010)29.
76. A.Kraft, *Int. J. Electrochem. Sci.,* 2(2007)355.
77. M. M. Antonijevic, M. B. Petrovic, *Int. J. Electrochem. Sci.,* 3(2008)1-28.
78. M. R. Ganjali, Z. Memari, F. Faridbod and P. Norouzi, *Int. J. Electrochem. Sci.,* 3(2008)1169.
79. M. R. Ganjali, R. Nemati, F. Faridbod, P. Norouzi and F. Darviche, *Int. J. Electrochem.Sci.,*3(2008)1288.
80. R. K. Mahajan and P. Sood, *Int. J. Electrochem. Sci.,* 2 (2007) 832.
81. M. R. Ganjali, M. Tavakoli, F. Faridbod, S. Riahi, P. Z Norouzi and M. S. Niassari, *Int. J.Electrochem. Sci.,* 3(2008)1559.
82. A.S. Al Attas, *Int. J. Electrochem. Sci.,* 4(2009)9.
83. A.S. Al Attas, *Int. J. Electrochem. Sci.,* 4(2009)20.
84. N. M.H. Rizk, S. S. Abbas, F. A. EL-Sayed and A. Abo-Bakr, *Int. J. Electrochem. Sci.,* 4(2009)396.
85. M. R. Ganjali, H. Ganjali,B. Larijani and P. Norouzi, *Int. J. Electrochem. Sci.,* 4 (2009) 914.
86. V. K. Gupta, R. N. Goyal, A. K. Jain and R. A. Sharma, *Electrochim. Acta, 54*(2009) 3218.
87. V. K. Gupta, R. N. Goyal and R. A. Sharma, *Anal. Chim. Acta,* 647(2009)66-71.
88. V. K. Gupta, M. K. Pal and A.K. Singh, *Talanta*, 79(2009) 528.
89. V. K. Gupta, R. N. Goyal, M. K. Pal and R. A. Sharma, *Anal. Chim. Acta,* 653(2009)161.
90. V. K. Gupta, R. Mangla, U. Khurana and P. Kumar, *Electroanalysis,* 11 (1999) 573.
91. V. K. Gupta, A. K. Jain and G. Maheshwari, *Talanta*, 72 (2007)1469.
92. V.K.Gupta, A. K. Jain and P. Kumar, *Sens. Actuat. B,* 120 (2006) 259.
93. V. K. Gupta, A. K. Jain and G. Maheshwari, *Int. J. Electrochem. Sci.,* 2 (2007) 102.
94. V. K. Gupta, R. N. Goyal, and R. A. Sharma, *Int. J. Electrochem. Sci.,* 4 (2009) 156**.**
95. A.K. Jain, V.K. Gupta, S. Radi, L.P. Singh and J.R. Raisoni, *Electrochim. Acta*, 51(2006) 2547.
96. A.J. Hamdan*, Int. J. Electrochem. Sci.,* 5 (2010) 215.
97. F. Faridbod, M. R. Ganjali and P.Norouzi, *Int. J. Electrochem. Sci.,* 4 (2009) 1679.
98. V. K. Gupta, R. Jain and M. K. Pal, *Int. J. Electrochem. Sci.,* 5 (2010) 1164.
99. W. Zhang, L. Jenny and U.E. Spichiger, *Anal. Sci.* 16 (2000) 11.

100. P.C. Meier, D. Ammann, W.E. Morf and W. Simon, Liquid-membrane ion-selective electrodes and their biomedical applications, in: J. Koryta (Eds.), Medical & Biological Application of Electrochemical Devices, Wiley, 1980, pp. 19-22.
101. O. Dinten, U.E. Spichiger, N. Chaniotakis, P. Gehrig, B. Rusterholz, W.E. Morf and W. Simon, *Anal. Chem.,* 63 (1991) 596-603.
102. S. Jadav and E. Bakker, *Anal. Chem.,* 73 (2001) 80-90.
103. M.A.A. Perez, L.P. Martin, J.C. Quintana and M. Y. Pedram, *Sens. Actuators B,* 89 (2003)262.
104. V. K. Gupta, R. Prasad and A. Kumar, *J. Appl. Electrochem.,* 33(2003)381.
105. V. K. Gupta, R. Prasad, A. Kumar, *Talanta,* 60(2003)149.
106. V. K. Gupta, M.M. Antonijevic, S.Chandra and S.Agarwal, *Sensors,* 2 (2002) 233.
107. A.K. Jain, V. K. Gupta, L. P. Singh and U. Khurana, *Analyst,* 122(1997) 583.
108. V. K. Gupta, A. K. Singh and B. Gupta, *Anal. Chim. Acta,* 583(2) (2007) 340.
109. I.Ali and V. K. Gupta, *Nature Protocols,* 1(6) (2007) 2661-2667**.**
110. V.K Gupta, A.Mittal, V.Gajbe and J. Mittal, *J. Coll. Int. Sci*., 319(2008) 30-39.
111. V.K Gupta, A.Mittal, R.Jain, M. Mathur and S. Sikarwar, *J. Coll. Int. Sci*., 303(2006)80-86.
112. V.K. Gupta, *The Arabian J. Sci. Engg. A-Science,*35(2A), (2010)7-25.
113. V.K. Gupta, P.J.M. Carrott, M.M.L. Ribeiro Carrott, *Critical Reviews in Environmental Science and Technology,* 39(2009).783–842 .
114. V. K. Gupta, M. Al Khayat, A.K. Singh and Manoj. K. Pal, *Anal.Chim. Acta,* 634(2009)36-43.
115. S. K. Srivastava, V. K. Gupta, M. K. Dwivedi and S. Jain, *Analytical Proceedings including Analytical Communications* ,32 (1995) 21-23.
116. A.K. Jain, V. K. Gupta, L. P. Singh, P. Srivastava and J. R. Raisoni, *Talanta,* 65(2005)716.
117. V.K. Gupta, Shilpi Agarwal and Barkha Singhal, *Combinatorial Chemistry & High Throughput Screening*, 14(4)(2011) 284-302.
118. V. K. Gupta, B. Sethi, N. Upadhyay, S. Kumar, R. Singh, L. P. Singh, *Int. J. Electrochem. Sci*., 6(30(2011) 650 - 663.
119. P. Norouzi, V. K. Gupta, F. Faridbod, B. Larijani, M. R. Ganjali, *Anal. Chem*., 83(5), 2011) 1564–1570.
120. V. K. Gupta, A. K. Jain, M. K. Pal, Shilpi Agarwal and A. K. Bharti, *Anal. Methods*, 3 (2011)334 – 342.
121. V. K. Gupta, Rajeev Jain, Milan M. Antonijevic, M. N. Siddiqui. A. Dwivedi , Shilpi Agarwal and R. Mishra, *Int. J. Electrochem. Sci*., 6 (2011) 37 – 51.
122. V. K. Gupta, A. J. Hamdan, R. Jain, S. Agarwal, A. K. Bharti, *Anal. Chim. Acta*, 681(2010)27-32
123. H. Khani, M. K. Rofouei, P. Arab, V. K. Gupta, Z. Vafaei, *J. Hazardous Materials*, 183 (2010)402-409.
124. V. K. Gupta, Manoj K. Pal and R. A. Sharma, *Talanta* 82(2010) 1136-1142.
125. P. Norouzi, G. R. Nabi Bidhendi, M.R. Ganjali, A. Sepehri, M. Ghorbani, *Microchim Acta* 152 (2005)123.
126. V.K. Gupta and I. Ali, *Environ. Sci. Technol.,* 42(2008)766-770.