Sensors **2007**, *7*, 400-409

sensors

ISSN 1424-8220

www.mdpi.org/sensors

Full Research Paper

A New Tiapride Selective Electrode and Its Clinical Application

Joaquín A. Ortuño, Vicente Ródenas, Mª Soledad García, Mª Isabel Albero and Concepción Sánchez-Pedreño *

Department of Analytical Chemistry, Faculty of Chemistry, University of Murcia, 30071 Murcia, Spain.

E-mail: jortuno@um.es, vicente.rodenas@carm.es, msgarcia@um.es, mialbero@um.es, spedreno@um.es

* Author to whom correspondence should be addressed.

Received: 29 January 2007 / Accepted:19 March 2007 / Published: 27 March 2007

Abstract: The construction and electrochemical response characteristics of a poly(vinyl chloride) (PVC) membrane selective electrode for the determination of tiapride (TPD) are described. The sensing membrane comprised an ion-pair formed between the protonated drug and tetraphenylborate (TPB^-) in a plasticized PVC matrix. The influence of membrane composition on the electrode response was studied. The electrode showed a fast, stable and Nernstian response over a wide tiapride concentration range ($1x10^{-5}$-$1x10^{-2}$ M) with a slope of 57.5±0.2 mV dec^{-1} of concentration, a detection limit of $4.3x10^{-6}$±$2.5x10^{-7}$ M, a wide working pH range (2-8) and a fast response time (< 5 s). The electrode showed good selectivity towards tiapride with respect to some inorganic and organic compounds. The electrode has been applied to the determination of tiapride in human urine and iontophoresis solution.

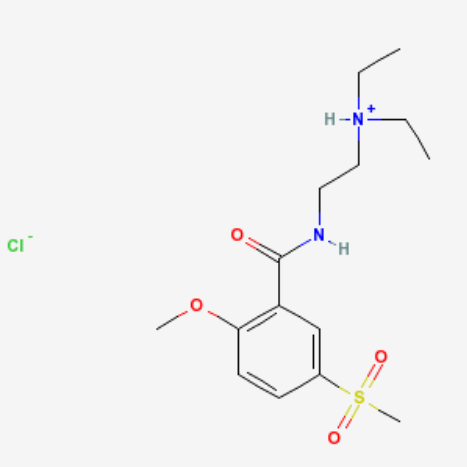
Keywords: Ion-selective electrode, tiapride determination; urine, iontophoresis solution.

---

1. Introduction

Substituted benzamides have been the first class of atypical anti-psychotics successfully employed both schizophrenia and depression. They are neuroleptics highly selective for dopamine receptors. Tiapride (N-[2-(diethylamino) ethyl]-5-(methylsulfonil)-o-anisamide, Scheme 1) (TPD), is a substituted benzamide with antipsychotic properties. It acts as antagonist of the dopamine D_2 receptors, a property which distinguishes this compound from other anti-psychotic agents. This particular feature

may explain the very low incidence of side effects on the extrapyramidal system. In addition tiapride has anti-emetic, anti-dyskinetic and anti-hypertensive actions [1].



Scheme 1

A review of the literature revealed that several analytical methods have been described for its determination, including chromatographic [2-3], spectrophotometric [4-5], fluorimetric [6], FI-chemiluminometric [7], and polarografic [8] methods.

In recent years, potentiometric membrane ion-selective electrodes (IESs) have been used in pharmaceutical and biological analysis [9-16]. This is mainly due to their simple design, low cost, adequate selectivity, good accuracy, wide concentration range and applicability to coloured and turbid solutions. However, a through literature survey has revealed no methods involving selective electrodes for the determination of tiapride.

Therefore, the aim of this work was to develop a polymeric ion-selective electrode for tiapride determination and its application for determining this drug in urine and in an iontophoresis solution. Our scope of work is to develop sensors for point of care clinical analysis in the treatment of mentally ill patients.

2. Results and Discussion

2.1. Influence of membrane composition

Several membranes of the different compositions (Table 1) prepared as described in the Experimental were tested. Two plasticizers, NPOE and DOS, with very different dielectric constants were tested as membrane solvent. The calibration graphs obtained for the corresponding membranes, A and B respectively, are shown in Fig. 1.

Table 1. Composition of the membrane.

Membrane	Percentage (w/w) of components in membranes			
	PVC	NPOE	DOS	TPD-TPB
A	33.0	66.0	–	1.0
B	33.0	–	66.0	1.0
C	32.3	64.7	–	3.0

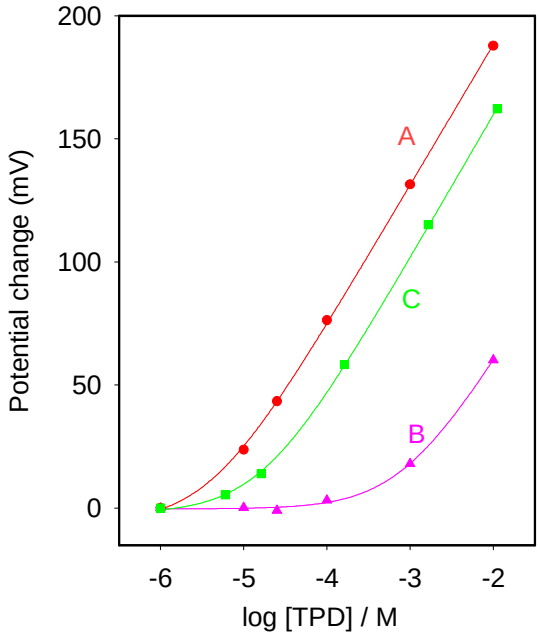


Figure 1: Calibration graphs for membranes A, B and C.

As can be seen the membrane plasticized with NPOE presented better response and a lower detection limit. The detection limit of ISEs with dissolved ion exchangers is controlled by the analyte ion concentration present in the solution as a result of the distribution equilibrium for the ion pair between the membrane and the solution [17]. The solubility of the ion-pair in the organic solvent generally increases as the polarity and dielectric constant increases [18], which explain the lower detection limit obtained with NPOE as plasticizer which was selected for further studies.

Two different TPD-TPB concentrations, 1.0 and 3.0 %, in the membrane were tested, (membranes A and C respectively). The corresponding calibration graphs are shown in Fig.1. As can be seen, the slopes in the linear range are very similar but the detection limit of membrane A (4.3×10^{-6} M) is lower than that of membrane B (1.7×10^{-5} M), which is due to the lower TPD concentration in the aqueous solution as a result of the distribution equilibrium of the TPD-TPB. Taking all these results into account, membrane A was selected for further studies.

2.2 Influence of pH

The effect of pH on the electrode potential at various tiapride concentration in the range 2.5x10^{-5}-1x10^{-2} M, was studied. The pH was varied by adding HCl or NaOH and the results are shown in Fig. 2. As can be seen, the electrode potential was independent of pH in the range 2-8 for all the tiapride concentrations assayed, and in this range the electrode can be safely used for tiapride determination. At higher pH values, the potential decreased due to the gradual increase in the concentration of the unprotonated form of the TPD. The pH of the tiapride working solutions prepared as described in *2.2*, is between 6-7 and therefore lies within the range of constant response.

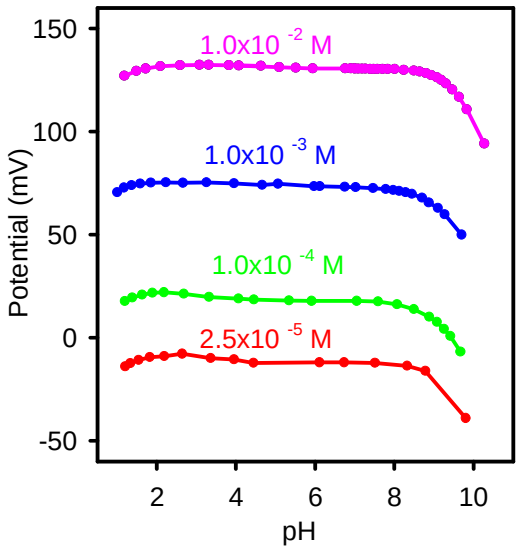


Figure 2: Influence of pH at different tiapride concentrations

2.3 Response characteristics

Ion-selective electrode characterization carried out with a mathematical and computational program has been shown to be very useful for the determination of detection limits and selectivity constants, among others [19]. Non-linear curve fitting using available common software was used for the determination of the ISE characteristics [20]. The slope and the detection limit for tiapride of the selected electrode were determined by fitting calibration data to equation (1):

$$E = E_i^0 + S\log(LOD + C_{TPD}) \quad (1)$$

The calibration parameters were evaluated from repeatedly making calibration graphs between tiapride 1x10^{-6} and 1x10^{-2} M. The calibration parameters obtained are shown in Table 2. As can be seen, a near-Nernstian response within a three decade concentration range, with low detection limit and good calibration reproducibility, was obtained.

The repeatability and reproducibility of the calibration parameters were studied by making successive calibrations with the same membrane on the same day (n=5) and on different days (n=5). The results obtained for the LOD were $4.3x10^{-6} \pm 2.5x10^{-7}$ M and $6.1x10^{-6} \pm 4.2x10^{-7}$ M, respectively, and for the slopes, 57.5 ±0.2 and 57.4 ±0.3 mV per decade of concentration, respectively.

The response time was obtained from the dynamic potential response corresponding to tiapride concentration steps to obtain a 10-times more concentrated solution. The values obtained for different tiapride concentrations are included in Table 2, and show a very rapid response even for low concentrations.

Table 2. Response characteristics of the tiapride-selective electrode.

*Slope (mV per dec) ± S.D.	57.5 ± 0.2
Linear range (M)	$1x10^{-5}$ to $1x10^{-2}$
*Detection limit (M) ± S.D	$4.3 \times 10^{-6} \pm 2.5x10^{-7}$
Response time (s) 10^{-6}-10^{-2} (M)	$t_{95\%} \leq 5$
Working pH range (10^{-6}-10^{-2} M)	2 – 8
Lifetime (months)	>3

*Mean of five calibrations

The tiapride selective electrode worked for at least 3 months, during which time no appreciable change in the calibration characteristics or response time was observed.

2.4 Selectivity

Selectivity coefficients for different ions with respect to tiapride were determined by the activity ratio method [21], in which the selectivity coefficient is measured as the ratio of ion activities or concentrations that generate the same membrane potential when measured in a separate solution type experiment. A tiapride calibration graph carried out in water was used to calculate the concentration of tiapride (C_{TPD}) that corresponds to the potential observed for a certain concentration (C_j) of interfering ion. The selectivity coefficients were calculated as the ratio of these concentrations, $K_{TPD, j} = C_{TPD} / C_j$, where j is the interfering ion.

The resulting selectivity coefficients are shown in Table 3. Ca^{2+}, Mg^{2+}, lactose, glucose and sucrose did not produce any change in the potential up to $1x10^{-1}$M with respect to the blank. When the concentration of these species was substantially increased, the potential decreased (i.e. it provided negative interference).

Table 3. Selectivity coefficients.

Interfering species	Log $K_{TPD, j}$	[j] / M assayed
Na^+	-5.16	2
K^+	-4.75	1
Ca^{2+}, Mg^{2+}	No interference up to 1x 10^{-1} M	
Lactose, glucose, sucrose	No interference up to 1x 10^{-1} M	

2.5 Applications

The new tiapride-selective electrode was satisfactorily applied to the determination of tiapride in human urine and in HEPES saline buffer, a medium usually used for iontophoresis samples.

To determine TPD in human urine a previous study of the possible interference from the sample matrix was carried by making calibration graphs applying the procedure described in *3*.5 to spiked blank urine sample with different amounts of TPD. The results obtained showed lower slopes and lower recoveries then those obtained in the absence of urine matrix. Therefore, the prior TPD extraction step described in *3.6*, was carried out. Known amounts of TPD were added to urine samples and the analyses were performed following the procedure described in *3.6*. The results obtained are summarized in Table 4. Good recoveries were obtained.

Table 4. Determination of tiapride hydrochlorhide in different samples.

Sample	Tiapride (mg L^{-1})*		Recovery
	Added	Found	%
Human urine	3.42	3.50±0.02	102.3
	34.2	33.8±1.9	99.0
HEPES saline	0.34	0.36±0.03	105.9
	2.06	2.09±0.10	101.4
	5.42	5.24±0.25	96.6
	22.2	21.6±0.7	97.7
	54.7	54.9±2.7	100.4

* Mean ± SD (n=3)

Tiapride is rapidly and efficiently absorbed after oral administration and is eliminated principally by hepatic metabolism and subsequent urinary excretion. The normal dose for adults is 300-1200 mg/day. Tiapride undergoes only limited metabolism, nearly 70 % of an orally administered dose is excreted unchanged in urine. The concentration of TPD in urine passed during 24h lies within the range of TPD determination of the potentiometric method proposed.

Iontophoresis is the application of a small electric current to enhance the transport of both charged and neutral compounds across the skin [22]. HEPES buffer (derived from N-(2-hydroxyethyl)-

piperazine-N'-2-ethanesulfonic acid) is a physiological buffer used in clinical analysis as an alternative to conventional phosphate buffer and HEPES saline buffer is a collector vehicle frequently used in iontophoresis reverse. To determination of tiapride in this medium, known amounts of TPD were added to HEPES saline buffer samples at pH 7.5 and the analyses were performed by direct potentiometry following the procedure described in *3.7.* The results obtained are summarized in Table 4. Good recoveries were obtained.

3. Experimental Section

3.1. Apparatus

Potentials were measured with an Orion 960 Autochemistry System, the recorder output of which was connected to a personal computer via a DGH Corporation 1121 module analogue-to-digital converter (Manchester, UK). An Orion 90-02 double junction silver-silver chloride reference electrode containing 0.1 M KCl solution in the outer compartment was used.

3.2. Reagents and solutions

Polyvinyl chloride (PVC) of high molecular mass, 2-nitrophenyl octyl ether (NPOE), bis (2-ethylhexyl) sebacate (DOS) and tetrahydrofuran (THF) were Selectophore products from Fluka. Tiapride hydrochloride powder, sodium tetraphenylborate (NaTPB), sodium salt of (N-[2-hydroxyethyl] piperazine-N´-[2-ethane suphonic acid]) (HEPES) and sodium sulphate were from Sigma. Al other reagents used were of analytical reagent grade, and nanopure water (18-MΩ) prepared with a Milli-Q (Millipore) system was used throughout.

3.2.1. Standard tiapride hydrochloride solution $5x10^{-1}$ M

This was prepared by dissolving 4.5613 g of pure drug (Sigma) in 25.0 ml of water. Working solutions ($1x10^{-2}$ to $1x10^{-6}$ M) were prepared by appropriate serial dilutions with $1x10^{-1}$ M sodium sulphate.

3.2.2. HEPES- buffered saline solution of pH 7.4

This was prepared by dissolving 1.63 g of N-2 hydroxiethylpiperazine-N´-2 ethanosulfonate sodium (HEPES sodium salt) and 1.94 g of NaCl in 250 ml of water. The pH 7.4 was adjusted with diluted HCl. The resulting solution contained $2.5x10^{-2}$ M HEPES and $1.3x10^{-1}$ M NaCl.

3.3 Ion pair preparation

The tiapride tetraphenylborate (TPD-TPB) ion pair was prepared by reacting 10 ml of $1x10^{-2}$ M tiapride hydrochloride solution with an equal volume of 1×10^{-2} M sodium tetraphenylborate solution. The mixture was filtered through a porous sintered glass crucible. The residue was washed first with distilled water until no chloride ion was detected in the washing solution, then with ether and finally dried at room temperature.

3.4 Construction and conditioning of the electrode

The membranes were prepared by dissolving 3.1 or 9.3 mg of TPD-TPB ion pair, 100 mg PVC and 200 mg oh the plasticizer (NPOE o DOS) in 3 ml of tetrahydrofuran. This solution was poured into a Fluka glass ring (inner diameter 28 mm, height 30 mm) on a Fluka glass plate. The solution was allowed to evaporate overnight. A 7 mm diameter piece was cut out with a Fluka punch for ion-selective membranes and incorporated into a Fluka electrode body ISE containing $1x10^{-2}$ M sodium chloride and $1x10^{-3}$ M tiapride and saturated with excess AgCl as internal filling solution. The compositions of the different membranes assayed are shown in Table 1.

The e1ectrodes were conditioned by soaking with constant stirring in a solution containing $1x10^{-3}$ M tiapride in $1x10^{-1}$ M Na_2SO_4 until the electrode gave a constant potential. When not in use, the electrode was kept immersed in the same dissolution.

3.5 General procedure (calibration of the electrode)

Standard tiapride solutions of $1x10^{-6}$-$1x10^{-2}$ M in $1x10^{-1}$ M Na_2SO_4 were prepared. The tiapride-selective and reference electrodes were immersed and the potential of each sample solution was directly measured. The measured potentials were plotted versus logarithmic values of concentrations and the calibration parameters were calculated by fitting calibration data to the equation shown in *3.3*. For the dynamic response studies, the electrode was calibrated by injecting, while stirring, adequate small volumes of tiapride standard solution in 50 ml of $1x10^{-1}$ M Na_2SO_4 to obtain final concentrations in the range $1x10^{-6}$-$1x10^{-2}$ M.

3.6. Procedure for the determination of tiapride in human urine

In the absence of human urine samples containing tiapride, known amounts of tiapride hydrochloride were added to 50 ml of blank urine samples and the pH was adjusted to about 11 with 1.0 M NaOH before adding 10 ml of dichloromethane. The solution was shaken and the aqueous phase was discharged. The organic phase was shaken with 40 ml of $1x10^{-2}$ M HCl solution and the aqueous phase was separated for further use. The pH of this phase was adjusted to about 7 with 1.0 M NaOH and diluted to 50 ml with water in a calibration flask. The tiapride-selective and reference electrodes were immersed and the tiapride concentration was determined by the standard addition technique.

3.7 Procedure for the determination of tiapride in iontophoresis solution

Frequently, iontophoresis experiments are carried out using a buffer HEPES saline solution. In the absence of human iontophoresis samples containing tiapride, known amounts of tiapride hydrochloride were added to 50 ml of buffer HEPES saline solution of pH 7.5 to obtain final concentration in the range $1x10^{-6}$ -$1.5x10^{-4}$ M tiapride. The tiapride-selective and reference electrodes were immersed and the tiapride concentration was determined by direct potentiometry using a calibration graph prepared in the same medium as the sample.

4. Conclusions

The new ion selective electrode developed, based on cation transfer across a plasticized poly (vinyl chloride) (PVC) membrane that contained the ion-pair formed between the protonated tiapride and tetraphenyl borate provides a rapid, sensitive, precise and inexpensive method for direct potentiometric determination of tiapride. The proposed method allows the determination of tiapride, in human urine samples, within the physiological concentration range obtained after the usual therapeutic dose of tiapride has been administered, and in a collector vehicle frequently used in the iontophoresis reverse.

Acknowledgements

We gratefully acknowledge the Ministerio de Ciencia y Tecnología, Spain (Project CTQ 2005-06977 / BQU) and FEDER (Fondo Europeo de Desarrollo Regional) for the financial support of this work

References

1. Spano, R.F.; Trabucchi, M.; Corsini, G.U.; Gessa, G.L. Sulpiride and other benzamides,Experimental and Clinical Pharmacology, *Raven Press, New York,* **1979.**
2. Chiba, R.; Ogasawara, A.; Kubo, T.; Yamazaki, H.; Umino, M.; Ishizuka, Y.Direct determination of benzamides in serum by column-switching high-performance liquid chromatography, *Anal. Sci*. **2003**, *19*, 785-789.
3. Moon, Y.; Paek, I.B.; Kim, H.-H.; Ji, H.Y.; Lee, H.W.; Park, H.-G.; Lee, H.S. Determination of tiapride in human plasma using hydrophilic interaction liquid chromatography-tandem mass spectrometry, *Archives of Phamacol. Research*, **2004**, *27*, 901-905.
4. Radwan, M.F. Spectrophotometric determination of three benzamide antipsychotic in pure and pharmaceutical preparation using charge transfer complex, *Egypt. J. Biomed. Sci*., **2003**, *11*, 1-12.
5. Attia, K.A.; Abou-Seada, H.H.; Nassar, M.W.I.; Colorimetric determination of certain dopamine antagonists in pharmaceutical preparations, *Egypt. J. Biomed. Sci.*, **2003**, *12*, 199-208.
6. Buna, M.; Aaron, J.J.; Prognon, P.; Mahuzier, G. Effects of pH and solvent on the fluorescence properties of biomedically important benzamides. Application to determination in drugs and in human urine, *Analyst*, **1996**, *121*, 1551-1556.
7. Aly, F.A.; Alarfaj, N.A.; Alwarthan, A.A. Flow-injection chemiluminometric analysis of some benzamides by their sensitizing effect on the cerium-sulphite reaction, *Talanta* **2001**, *54*, 715-725.
8. Zeng, Y.; Cao, H. Electrochemical behaviour and adsorptive properties of tiapride, *Yingyong Huaxue Bianji Weiyuanhui,* **1997,** *14*, 108-110.
9. Bouklouze, A.; Elbouzekraoui, M.; Cherrah, Y.; Hassar, M.; Kauffmann, J.M. Potentiometric sensor for hydroxyzine determination, *Electroanalysis,* **2002,** *14*, 1369-1374.
10. Vytras, K. The use of ion-selective electrodes in the determination of drug substances, *J. Pharm. Biomed. Anal.*, **1989,** *7*, 789-812.
11. Kimbeni Malongo, T.; Blankert, B.; Kambu, O.; Amighi, K.; Nsangu, J.; Kauffmann, J.M.; Amodiaquine polymeric membrane electrode, *J. Pharm. Biomed. Anal.*, **2006,** *41*, 70-76.

12. Ghoreshi, S.M.; Behpour, M.; Nabi, M. A novel naphazoline-selective membrane sensor and its pharmaceutical applications, Sensors Actuat. B, **2006,** *113*, 963-969.
13. Sánchez-Pedreño, C.; Ortuño, J.A.; Hernández, J.; Perchlorate-selective polymeric membrane electrode based on a gold complex: application to water and urine analysis, *Anal. Chim. Acta*, **2000,** *415*, 159-164.
14. Hassan, S.S.M.; Mahmoud, W.H.; Elmosallamy, M.A.F.; Almarzooqi, M.H. Iron(II)-phthalocyanine as a novel recognition sensor for selective potentiometric determination of diclofenac and warfarin drugs, *J. Pharm. Biomed. Anal.*, **2005,** *39*, 315-321
15. Ueda, K.; Yonemoto, R.; Komagoe, K.; Masuda, K.; Hanioka, N.; Narimatsu, S.; Katsu, T. Tris(2-ethylhexyl)phosphine oxide as an effective solvent mediator for constructing a serotonin.selective membrane electrode, *Anal. Chim. Acta*, **2006,** *565*, 36-41
16. Badawy, S.S.; Issa, Y.M.; Mutair, A.A. PVC membrane ion-selective electrodes for the determination of hyoscyamine in pure solution and in pharamaceutical preparations under batch and flow modes, *J. Pharm. Biomed. Anal.*, **2005,** *39*, 117-124
17. Koryta, J.; Stulik, K. *Ion-Selective Electrodes*, 2nd Edition, Cambridge, **1983**, p.31.
18. Senike, T.; Hasegawa, Y. *Solvent Extraction Chemistry Fundamentals and Applications*, Marcel Dekker, New York, **1977,** p.149.
19. Janata, J.; Josowicz, M.; Vanysek, P.; DeVaney, D.M. Chemical Sensors. *Anal. Chem*. **1998,** *70*, 179R-208R.
20. Kane, P.; Diamond, D. Determination of ion-selective electrode characteristics by non-linear curve fitting**.** *Talanta* **1997,** *44*, 1847-1858.
21. Glazier, A.; Arnold, M.A. Selectivity of membrane electrodes based on derivatives of dibenzyltin dichloride, *Anal. Chem.* **1991,** *63*, 754-759.
22. Delgado-Charro, M.B.; Guy, R.H. Transdermal iontophoresis for controlled drug delivery and non-invasive monitoring, *S.T.P. Pharm. Sci*. **2001,***11*, 403-414.