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A Novel Potentiometric Sensor for Determination of Tramadol Hydrochloride in Biological Samples

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Abstract- A unique potentiometric ion-selective PVC membrane sensor was developed based on tramadol-(tetraphenylborate) as the sensing element, tetraphenylborate as an additive, and dibutyl phthalate (DBP) as the plasticizer solvent for the measurement of Tramadol in pharmaceutical preparations. Different parameters such as the electrode conditioning time, the effect of electrode materials, and the impact of pH solution on the electrode performance were evaluated. The electrode showed a Nernstian slope of 59.5±0.4 mV/decade for tramadol ions. The potentiometric sensor reported here has been optimized to provide excellent analytical performance with a linear in the concentration spectrum 1.0×10^{-7}-1.0×10^{-1} mol L^{-1} and a limit of quantification of 7.0×10^{-8} mol L^{-1}. The sensor has a reaction time of 5 seconds and can be utilized in the pH range of 2.0 to 7.0. With a mean relative standard deviation of less than 2%, the approach is effective and reliable. The current electrode would be used to detect tramadol hydrochloride in biological samples with excellent results.

Keywords- PVC membrane; Tramadol hydrochloride; Ion-selective electrode; Potentiometric sensor; Potentiometry

1. INTRODUCTION

In recent years, the use of potentiometric membrane sensors in various fields, including medicine, industry, agriculture, pharmacy, etc., in terms of their clearness, cheapness, High sensitivity, and precision compared to other methods of decomposition chemistry have received much attention. Most of the experiments performed in medical diagnostic laboratories are using these electrodes are perform so it is estimated that more than one billion measurements by these sensors [1,2]. Extensive development in field ion-selective electrodes has been carried out since the time introduced membranes Containing polymer solvents. Polyvinyl chloride is very fast as one of these solvents has been accepted and is still used as a standard substrate for electrodes Ion-selective has been used [3-6]. Another advantage of these electrodes is that they can be directly used to measure tramadol in different samples such as blood serum and drug samples in the presence of other interference species and without the need to separate them, with good accuracy and precision.

Tramadol hydrochloride, (1RS,2RS) [(dimethylamino)methyl]-2-[(dimethylamino) methyl], a synthetic codeine analogue, is -1-(3-methoxyphenyl) cyclohexanol HCl (Figure 1). Tramadol hydrochloride has central analgesic qualities, functioning on certain opioid receptors in the same way that opioids like morphine and codeine do. Tramadol is an agonist for opioid receptors (quasi-opioids) as well as a codeine analog. Although the effect of tramadol is not fully understood, it has shown that by binding the major molecule of the drug to opioid receptors, the reuptake of norepinephrine and serotonin is poorly inhibited and is absorbed rapidly and almost completely. Tramadol is a white, crystalline powder that melts at 179°C. This substance is soluble in water and quickly absorbed by the body in any form, whether swallowed or suppository [7,8].

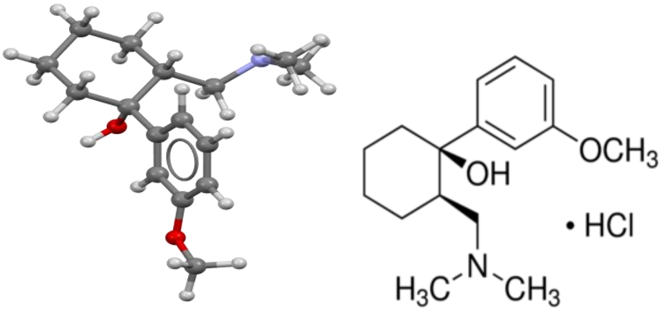

Figure 1. Tramadol's chemical structure as determined by hydrochloride

In recent years, some instrumental methods have been measured this drug at low concentrations and in different environments. Some techniques based on HPLC [9], GC with flame ionization detection [10,11]. Spectrophotometry [12], potentiometry [13-19] are used for determination of tramadol. The limitations of these methods are lengthy sample preparation processes, very high costs, sophisticated tools, well-equipped laboratories, and special training. Potentiometric ISEs with PVC membrane electrodes have been reported in pharmaceuticals in a few cases.

The ion-association of tramadol hydrochloride with dibutyl phthalate as a plasticizer is discussed in this paper, as well as the characteristics and analytical use of a unique tramadol ion-selective sensor. This study's stated electrode has a broad concentration spectrum, low detection limits, an excellent Nernstian slope, and strong discrimination across a broad range of other ions, and it was effectively used to measure tramadol hydrochloride in pharmaceutical tablet formulations and urine samples.

2. EXPERIMENTAL SECTION

2.1. Chemicals

The pure form of tramadol hydrochloride (C16H25NO2, MW=263.381g/mol) used in the present work supplied by Bakhtar Bioshimi Pharmaceutical Company (Kermanshah, Iran). Tramadol hydrochloride TDCl and tablets prepared from different local drug stores. All chemicals used of analytical or pharmaceutical grade, and the solutions prepared in double-distilled water. 2-Nitrophenyloctyl ether (2-NPOE), dioctyl phthalate (DOP), Bis-(1-butyl pentyl) Adipate (BBPA), Bis-(2-ethyl hexyl) Sebacate (BEHS), dioctyl sebacate (DOS), Dibutyl phthalate (DBP), tri-butyl phosphate (TBP), tris-2-ethyl-hexyl phosphate (TOP), NaTPB, high relative molecular weight PVC, and tetrahydrofuran (THF) were gathered from Fluka, or Merck and used as received.

2.2. Apparatus

Electrochemical Potentiometric evaluations were done with a 250 pH/mV metre with a resolution of ±0.1 mV at Zag Chimi, Iran, using the below setup:

$$\text{Ag} \mid \text{AgCl} \mid \text{internal solution (KCl 3.0 mol L}^{-1}\text{ TD cation } 1.0 \times 10^{-1}\text{ mol L}^{-1}) \mid$$
$$\text{PVC membrane} \mid \text{test solution} \parallel \text{SCE}$$

During stirring settings at room temperature (25.0±1.0°C), a saturated calomel electrode (SCE, Azar electrode, Iran) was utilized as an external standard electrode. Through serial dilution, the performance of electrodes is assessed on EMFs of TD solutions with concentration ranges of 10^{-8}–10^{-1} mol L^{-1}. Each solution's potential was measured and shown as a logarithmic function of TD cation activity after it was shaken and assessed.

2.3. The Ion-Pair Compound's Formulation

About 25 mL of 0.01 M tramadol hydrochloride was combined with 25 mL of 0.01 M tetraphenylborate solution under shaking to make the ion-pair product tramadol-tetraphenylborate (TD-TPB). Water was washed and dried at 60°C after precipitation and filtration.

2.4. Electrode preparation

To prepare a tramadol-sensitive sensor, first mix some PVC, tramadol tetraphenyl borate ion pair and various emulsifiers, DBP, DOS, TEPH, BA, DEPH, BBPA, DOP, 2-NPOE, DOPH DBS, DBBPH, and mixed solvent. About 3-5 ml THF was added, and the mixture was stirred for 15 minutes to obtain a uniform solution. A total mass of 100 mg of PVC, plasticizer, and membrane additive was diluted in around 5 mL THF. The ionophore (TDTPB) complex was added to this combination, and the solution was thoroughly stirred. Table 1 shows the various membrane compositions. The ideal membrane mixture was 30.0 wt.% PVC, 64.0 weight percent DBP, and 6.0 wt.% ionophore.

The mixture was then placed into a tiny flat-bottom dish with a diameter of 2 cm, covered with filter paper, and left to evaporate at room temperature. A nontransparent membrane was generated by dipping a Pyrex tube (5 mm i.d.) into the mixture for around 10 seconds. The tube was then ejected from the mixture and allowed to cool for 12 hours. The internal solution 1.0×10^{-1} mol L^{-1} TD^{+} has been added to the tube. The first conditioning period lasted about 12 hours, with subsequent conditioning periods lasting 20 to 30 minutes. As an internal reference electrode, a silver/silver chloride electrode has been utilized. 1×10^{-1} mol L^{-1} tramadol solutions were dissolved in water and dissolved in a 100 ml volumetric flask. Continuous dilution was used to make various solutions in the concentration range of 1×10^{-8}-1×10^{-1} mol L^{-1}. Water was twice distilled in all of the cases.

3. RESULTS AND DISCUSSION

3.1. Membrane ingredients optimization

Considering the change in percentage of the membrane components on the response of electrodes, optimization of the detail is essential. The concentration range of 7×10^{-7}-1×10^{-1} mol L^{-1} shows a linear response with a slope of 59.5 mV per decade of change in concentration.

The nature of the ionophore utilized and the qualities of the plasticizer were found to have a considerable impact on the specificity and sensitivity attained for a given ion-selective electrode, according to the research. After testing six solvents (2-NPHOE, DOP, DBP, DOPH, DOS, and DBBPH), it was revealed that DBP, with moderate volatility, solubility in lipid, molecular weight, and low dielectric constant, produced the best outcomes, as shown in Table 1 and Figure 2A. As a result, DBP was selected as a promising plasticizer for further investigation. Finally, the electrode with a membrane composition of no. 5 was selected as a final electrode for further experiments. The calibration of the sensor with membrane composition no. 5 is shown in Figure 2B.

Table 1. Components for membrane ingredients optimization

Response Time (s)	Linear Range (M)	Slope (mV/decade)	Plasticizer (%W)	Ion-Pair (%W)	PVC (%W)	Electrode No.
10	6.0×10^{-5} - 1.0×10^{-1}	24.8 ± 1.15	63(DBS)	6	31	1
10	1.0×10^{-5} - 1.0×10^{-1}	23.0 ±1.10	61(DBS)	6	33	2
10	1.0×10^{-5} - 1.0×10^{-1}	30.70±0.84	60(DOP)	7	33	3
10	1.0×10^{-6} - 1.0×10^{-1}	54.6±0.2	60(DOS)	8	32	4
5	7.0×10^{-7} - 1.0×10^{-1}	59.5±0.4	64(DBP)	6	30	5
10	1.0×10^{-6} - 1.0×10^{-1}	62.1±0.16	62(DOP)	6	32	6
15	1.0×10^{-7} - 1.0×10^{-1}	46.7±0.2	60(DOP)	7	33	7
15	1.0×10^{-6} - 1.0×10^{-1}	60.9±0.15	61(DOP)	7	32	8
10	1.0×10^{-6} - 1.0×10^{-1}	56.2±0.16	59(DBP)	7	34	9
15	3.0×10^{-5} - 1.0×10^{-1}	48.8±0.5	62(DBBPH)	7	31	10
10	1.0×10^{-6} - 1.0×10^{-1}	57.9±0.17	61(2-NPOE)	7	32	11
10	1.0×10^{-7} - 1.0×10^{-1}	52.9±0.2	61(DEPH)	8	31	12
15	1.0×10^{-6} - 1.0×10^{-1}	49.5±0.2	64(DOP)	6	30	13
10	1.0×10^{-5} - 1.0×10^{-1}	46.9±0.2	65(DOP)	5	30	14
10	1.0×10^{-6} - 1.0×10^{-1}	42.9±0.2	63(DOP)	5	32	15
10	1.0×10^{-5} - 1.0×10^{-1}	43.0±0.3	65(DOP)	0	35	16
10	1.0×10^{-6} - 1.0×10^{-1}	48.9±0.15	62(DOPH)	4	34	17
10	1.0×10^{-6} - 1.0×10^{-1}	50.9±0.2	64(TEPH)	5	31	18

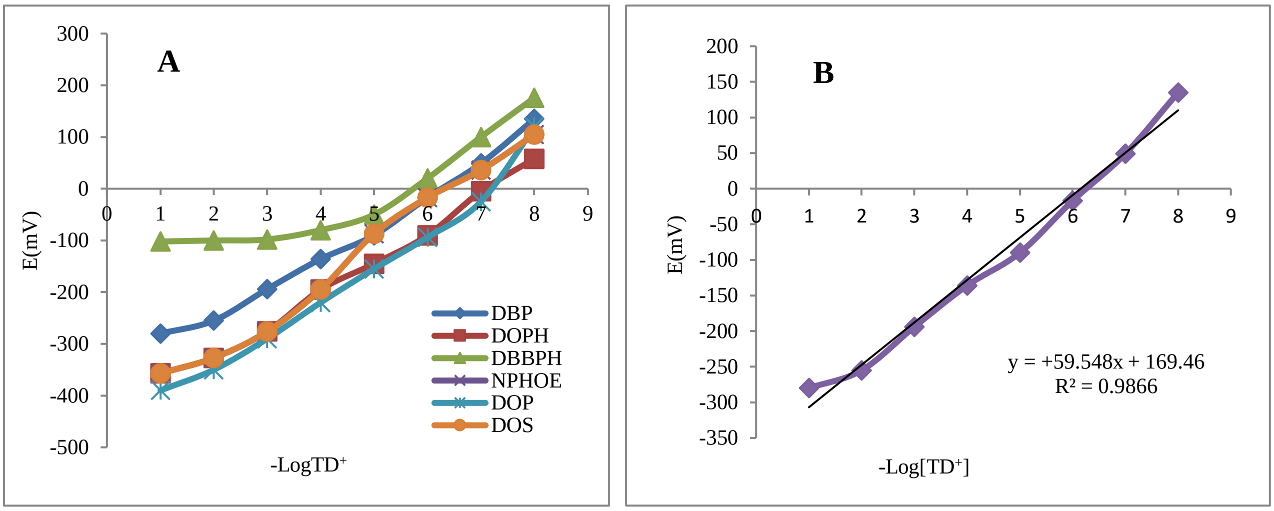

Figure 2. A) Varied plasticizers have different effects on the TD electrode (No.5) responsiveness; B) Calibration graph and detection limit of tramadol hydrochloride with DBP as a plasticizer

3.2. The Influence of pH on Electrode Performance

The influence of pH on response electrode response in 1.0×10^{-3} and 1.0×10^{-4} mol L^{-1}

TD solution was investigated after the membrane components had been optimized. As needed, dilute HCl or NaOH were used to adjust the pH. Within the pH range of 2.0-7.0 indicated in fig4, the potentials remained constant. Low pH acid solutions include hydronium ions, which compete efficiently with TD drug ions due to their great mobility. Nonetheless, when the sensor pH was greater than 7.0, there was a nonlinear response with a modest increase in the potential. At high pH, the potential of forming a neutral complex between tramadol ions and hydroxide ions increases, and OH ions permeate the membrane, abstracting the proton from the medication and changing it to the free amine. The response potential is lowered because tramadol ions do not permeate through the membrane.

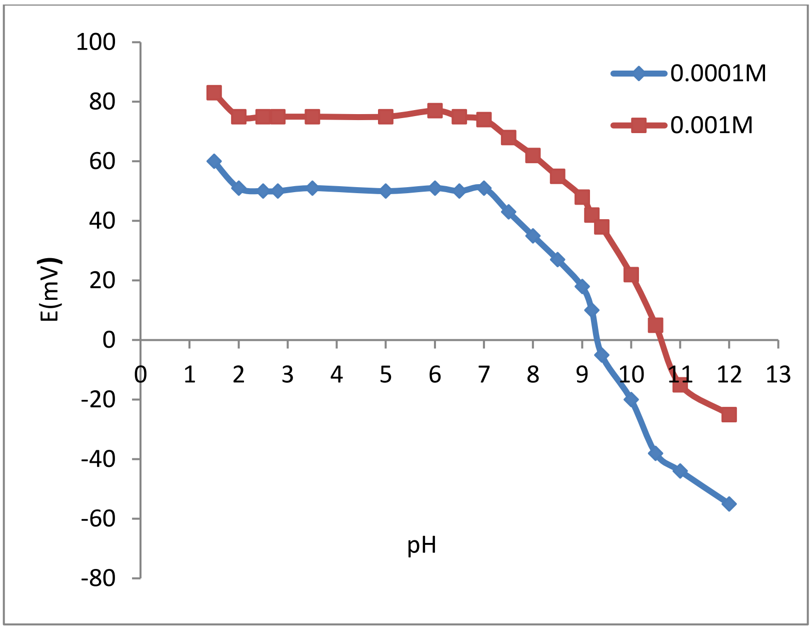

Figure 3. The pH of the test solution has an effect on the TD's potential response

3.3. Calculation of the Selectivity Index

The most crucial aspect of membrane electrodes is their selectivity. It determines whether or not the electrode can accurately measure the target ion in the presence of other interfering ions. The selectivity coefficient described the electrode's sensitivity to the target ion compared to other ions. The Matched Potential Method (MPM) [20] and the separated solution method were used to test the membrane electrode's selectivity in the presence of interfering species in solutions such as cations and biological components (SSM). When compared to other cations that are related to biological substances like Glycine, L-Histidine, D-Fructose, and Glucose, the electrode shows a high selectivity. As a result, these species have a negligible impact on tramadol hydrochloride measurement, and tramadol hydrochloride can be measured without interference in the presence of these species. Table 2 shows a summary of the findings.

To find the selectivity coefficient, use the following formula:

$$LogK_{Drug,J^{z+}}^{Pot} = \frac{E_2 - E_1}{S} + log[Drug] - log[J^{z+}]^{1/z} \quad (1)$$

In MPM, specific amounts of TDCl in the range of 1×10^{-4} to 1×10^{-5} M were applied to a TDCl reference solution, and the resulting potential change (E) was calculated. In another experiment, 1.0×10^{-1} M of the interfering ion (J) was introduced to an analogous reference solution in steps until the change in potential matched the E value. The following equation was then used to calculate the values:

$$LogK_{Drug,J^{z+}}^{Pot} = \frac{a_{Drug}}{a_J} \quad (2)$$

where the a_J is the activity of the added interferent.

Table 2. For the TD^+ selective electrode, the selectivity coefficient of various interfering ions

K(MPM)	K(SSM)	Interfering ions	K(MPM)	K(SSM)	Interfering ions
2.4×10^{-5}	5.0×10^{-5}	K^+	3.4×10^{-5}	1.9×10^{-5}	Na^+
6.2×10^{-4}	3.9×10^{-4}	Ni^+	5.6×10^{-4}	3.1×10^{-3}	Li^+
8.5×10^{-4}	4.2×10^{-3}	Ca^{2+}	5.1×10^{-4}	8.5×10^{-4}	Mg^{2+}
8.2×10^{-4}	3.4×10^{-3}	Cu^{2+}	7.4×10^{-4}	1.7×10^{-2}	Cr^{3+}
7.3×10^{-4}	1.7×10^{-3}	Co^{2+}	5.4×10^{-4}	3.8×10^{-4}	Zn^{2+}
5.3×10^{-5}	6.5×10^{-5}	Ascorbic acid	3.1×10^{-4}	2.4×10^{-3}	Fe^{3+}
-	2.8×10^{-5}	Glucose	-	1.1×10^{-6}	D-Fructose
7.8×10^{-3}	9.2×10^{-3}	Uric Acid	4.6×10^{-5}	7.9×10^{-5}	Sucrose
4.8×10^{-4}	4.1×10^{-3}	Ampicillin	4.8×10^{-4}	3.6×10^{-3}	Captopril
1.8×10^{-2}	5.3×10^{-3}	L-Histidine	5.0×10^{-4}	1.2×10^{-2}	Glycine

3.4. Electrode response time

To determine the electrode response time, the time it takes for the electrode potential to reach 90% of the final equilibrium value, potential measurements of solutions containing various tramadol concentrations were taken using solutions containing various TD^+ concentrations ranging from 1×10^{-6} to 1×10^{-2} mol L^{-1}. The curve of potential change *vs.* time was plotted (Figure 4). At greater concentrations, the electrode reaches equilibrium faster. This electrode has a reaction time of roughly 5 seconds. Figure 5 depicts the electrode's reaction time over the concentration range.

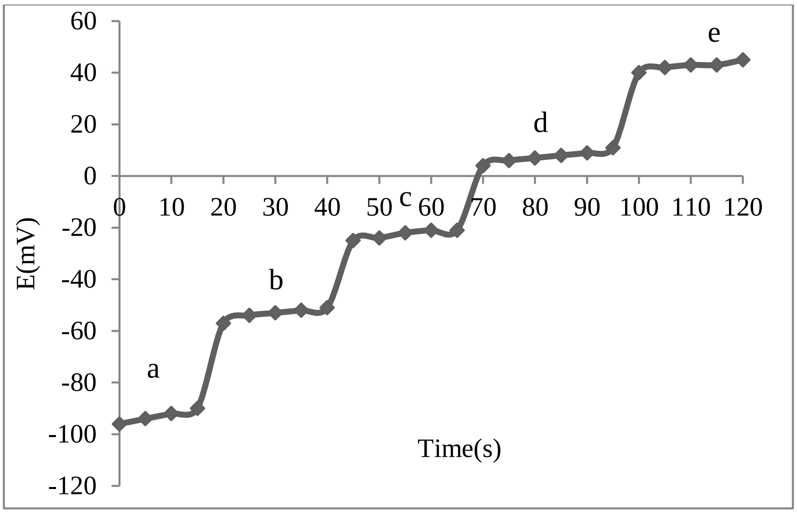

Figure 4. For the reaction of the TD electrode, a standard potential-time figure is shown. a) 1.0×10^{-6} mol L^{-1}; b) 1.0×10^{-5} mol L^{-1}; c) 1.0×10^{-4} mol L^{-1}; d) 1.0×10^{-3} mol L^{-1}; e)1.0×10^{-2} mol L^{-1}

3.5. Analytical applications

By using standard addition, the calibration curve, and potentiometric titration procedures, the suggested sensor was used to determine tramadol hydrochloride content in tablets and capsules. Table 3 shows that the findings are good due to the consistency and low standard deviation.

0.2000 g homogenized powder of tramadol tablets transferred into 100mL volumetric flask, and after complete dissolution in distilled water and using a magnetic stirrer, diluted to the marked line. A proposed electrode was used for determining tramadol content by calibration method [21]. Table 3 shows the findings of the tramadol quantity determination in different pharmaceutical preparations from a local pharmacy.

Table 3. Results of Tramadol Assay in Tablets by the Tramadol Membrane Sensor

Application Sample	**Stated Content (mg/tab)***	**Found (mg/tab)**
Tramal	100.0	100.2±0.4
tramad	100.0	100.2±0.5
byomadol	100.0	101.1±0.2
pyralgine	100.0	100.3±0.2

*Data obtained from five measurements

3.6. Measuring the spiked urine and milk samples

The novel sensor was used to recover tramadol from urine and milk samples to see if it might be used to determine drug concentrations in biological fluids. The samples (5mL urine and 10mL humanised cow milk) were spiked with tramadol hydrochloride and swirled for 5 minutes before being transferred to a 25 mL volumetric flask and finished with distilled water to yield 1.0×10^{-6} to 1.0×10^{-2} mol L^{-1} TD^+. For recovery and TD^+ determination, these mixtures are treated to either the usual addition or calibration graph methods [22].

Table 4. Determination of TD in urine and milk samples

Sample	Spiked (mol L^{-1})	Recovery (%)
Urine	1.0×10^{-3}	1.14 (±0.07) $\times10^{-3}$
Urine	1.0×10^{-4}	1.05 (±0.08) $\times10^{-4}$
Urine	1.0×10^{-5}	0.98 (±0.05) $\times10^{-5}$
Milk	1.0×10^{-3}	1.08 (±0.07) $\times10^{-3}$
Milk	1.0×10^{-4}	0.94 (±0.1) $\times10^{-4}$
Milk	1.0×10^{-5}	0.92 (±0.0.06) $\times10^{-5}$

4. CONCLUSION

The tramadol sensor was found to have great analytical efficiency features after a series of studies involving the use of TDTPB ion-pair complexes alongside certain plasticizers inside the membrane design. With a short response time (~5 s), a reduced detection limit of 7×10^{-7} mol L^{-1}, and pH-independent potential responses throughout the 2.0–7.0 range, it has demonstrated advanced efficiency. The sensor-enabled tramadol determination in pharmaceutical analysis has a high sensitivity.

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