



Highly Sensitive Electrochemical Sensor Determination Of Drug In Pharmaceutical Analysis



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MODIFIED carbon paste electrodes and screen printed electrodes were developed for the potentiometric determination of butenafine hydrochloride (BTFHC) in urine, serum and pharmaceutical preparations. Two modified carbon paste electrodes (electrodes A and C) and two modified screen printed electrodes (electrodes B and D) using tricresylphosphate (TCP) and o-nitrophenyloctyl ether (o-NPOE) plasticizers, respectively, were used. The electrodes showed a linear concentration range from 1.57×10^{-7} to 1.0×10^{-2} and 1.0×10^{-7} to 1.0×10^{-2} mol L⁻¹ with detection limit of 1.57×10^{-7} and 1.0×10^{-7} mol L⁻¹ for modified carbon paste (MCPEs; electrodes A and B) and modified screen printed (MSPEs; electrodes C and D), respectively. The calibration graphs have slope values of 56.95 ± 0.56 , 58.22 ± 0.78 , 56.18 ± 0.66 and 57.69 ± 0.39 mV decade⁻¹ for electrodes A, B, C and D, respectively. The response time was relatively quick in the whole concentration range and was found to be 12s for electrodes (A and B) and 7s for electrodes (C and D). The electrodes were found to have life time of 82, 99, 160 and 185 days without showing any deviations in a pH range of 5.0–9.0 and 4.0–9.0 for MCPEs (electrodes A and B) and MSPEs electrodes (C and D), respectively. The fabricated electrodes displayed good selectivity for BTFHC with respect to number of common foreign inorganic cations, sugar species, organic and glycine as the fillers that may be added to the pharmaceutical preparation or biological fluids. Fortunately, such materials mostly do not interfere. The potential application of the sensors (A-D) was accomplished as a result of good recoveries made from real samples fortified with BTFHC. Results indicated the proficiency of the sensors reliable for rapid, onsite monitoring of BTFHC in pharmaceutical preparation and in biological samples.

Keywords: Butenafine hydrochloride ion-selective electrodes, Modified carbon paste electrode, Modified screen-printed sensors, Pharmaceutical analysis, Potentiometric Determination.

Introduction

Butenafine hydrochloride (BTFHC) has the IUPAC name 1-(4-tert-butylphenyl)-N-methyl-N-(naphthalen-1-ylmethyl) methanamine hydrochloride Scheme 1 [1]. It is a synthetic benzyl amine antifungal [2] and belongs to the class of naphthalenes organic compounds [3].

It metabolized by liver and urine [1] where the metabolic pathways are methylation; dealkylation [4]. Many types of fungal germs (fungi) live harmlessly in the soil, on food, on our skin and in other places in the environment [5-7]. However, some types of fungi can thrive and multiply on the surface of the body, to cause infection of the skin, nails, mouth or vagina [6]. The most common

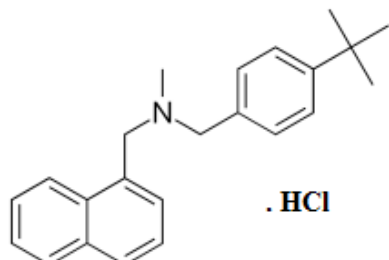
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fungi to cause skin infections are the tinea group of fungi [1]. Butenafine is indicated for the topical treatment of tinea as well as athlete's foot (*Tinea pedis*), ringworm (*Tinea corporis*) and jock itch (*Tinea cruris*) [8]. It also displays superior activity against *Candida* than terbinafine and naftifine.



Scheme 1: Chemical structure of butenafine hydrochloride.

In wide variety of applications, various ions in aqueous solutions were determined using ion-selective electrodes (ISEs) [9-11]. In comparison with other analytical methods, they are relatively inexpensive and simple to use [12-15]. Carbon-paste electrodes (CPEs) have several advantages of very low Ohmic resistance, low cost, very short response time, reproducibility of the preparation process, very simple, cheap and quick preparation process. Which provide the possibility of measurements on small volumes as well as ability for construction of a portable titration system for field titration of drug [16, 17]. These electrodes are commonly used for potentiometric measurements; however, they are also applicable in coulometry (potentiometry) [18, 19].

Screen-printed electrodes (SPEs) had low fabrication cost and disposable devices with short response times capable of performing routine electrochemical biosensing. Disposable screen-printed electrodes provide fast, sensitive detection and quantification of various analytes in complex samples derived by the medical diagnosis and food fields. In addition, the small size of these electrodes enables a very small volume of sample to be used for analysis [20-24].

The present work aims to develop modified carbon past and screen printed ion-selective electrodes for the determination of some drugs based on using β -cyclodextrine and multiwall carbon nanotube (MWCNT) ionophores. The purpose of this investigation was directed to describe sensitive, accurate and reproducible potentiometric method that can be applied to determine BTFHC drug under investigation in *Egypt. J. Chem.* **63**, No. 8 (2020)

pure, in different pharmaceutical preparations and biological fluid samples. The performance of the fabricated modified carbon paste and screen printed ion selective electrodes was optimized according to the ICH guidelines. The percent recovery, accuracy and precision, limit of detection and limit of quantifications were calculated.

Experimental

Apparatus

HANNA 211 pH meter and Thermo-Orion, model Orion 3 stars (USA) were used for potential and pH measurements. Double-junction Ag/AgCl (Metrohm 6.0726.100) reference electrode and digital burette were used. All glassware were washed carefully with bidistilled water and dried in the oven prior to their use in analysis.

Reagents and Chemicals

In this study analytical grade reagents and bidistilled water were used throughout all experiments. They included butenafine hydrochloride (BTFHC) which was provided by Egyptian Group for Pharmaceutical Industries. Tricresylphosphate (TCP) and *o*-nitrophenyloctyl ether were supplied from Alfa Aesar and Fluka, respectively, and were used for the preparation of the sensors. Dioctylphthalate (DOP), dibutylphthalate (DBP) and dioctylsebacate (DOS) were supplied from Sigma, Merck and Merck, respectively. Graphite powder (synthetic 1–2 μm) and polyvinyl chloride (PVC, relative high molecular weight) were supplied from Aldrich. β -Cyclodextrine (β -CD) and Multiwall carbon nanotube (MWCNT) were purchased from sigma and EPRI, respectively. Sodium tetraphenylborate (NaTPB) was purchased from Fluka. Cyclohexanone and acetone were supplied from Fluka (Switzerland). Hydrochloric acid and sodium hydroxide were supplied from BDH.

Interfering ions solutions

All solutions must be protected from light by keeping them in dark colored quickfit bottles during the whole work. Standard solution ($1.0 \times 10^3 \text{ mol L}^{-1}$) of sucrose, maltose, glycine, picric acid, glucose, fructose, lactose, urea, starch, chloride salts of chromium, calcium, ferric, sodium, potassium, zinc, cobalt, nickel, manganese, cadmium, aluminum and copper were used as interfering ions which prepared by dissolving the proper weights into 100 mL bidistilled water.

Butenafine hydrochloride solution

Stock BTFHC solution ($1.0 \times 10^{-2} \text{ mol L}^{-1}$) was prepared by dissolving the proper weight

of the drug (353.93 mg) into smaller amount of warmed distilled water, with stirring till the drug completely dissolved. The resulting solution was then made up to 100 mL with distilled water in a measuring flask.

Pharmaceutical samples

Tinearest cream (sample 1; Western Pharmaceutical Industries, El Obour City Avenue (1), Cairo, Egypt), Bravamax cream (sample 2; Chemipharm Pharmaceutical Industries, 6th October City, Cairo, Egypt) and Farbicure cream (sample 3; Farabi Pharmaceutical Industries, Abbas El-Akkad, Cairo, Egypt) were used. Each one gram contains 0.01 g BTFHC.

Electrodes preparation

Preparation of modified carbon paste electrodes (MCPEs)

A 480 mg pure graphite powder, 20 mg multiwall carbon nanotube and 5.0-15 mg of β -cyclodextrine (β -CD) and multiwall carbon nanotube (MWCT) ionophores are transferred to mortar and mixed well with plasticizer (0.2 mL of DOP, TCP, DBP, DOS or o-NPOE). The modified paste was filled in electrode body and kept in distilled water for 24 h before use. A fresh surface was obtained by gently pushing the stainless-steel screw forward and polishing the new carbon-paste surface with filter paper to obtain a shiny new surface when polished on a paper [25-28].

Preparation of the modified screen-printed electrodes (MSPEs)

Modified SPEs were printed in arrays of six couples of the working electrodes (each 5×35 mm) following the procedures previously described [27-30]. A polyvinyl chloride flexible sheet (0.2 mm) was used as a substrate which was not affected by the curing temperature or the ink solvent and easily cutted by scissors. The homemade printing ink was prepared by thoroughly mixing the cyclohexanone-acetone mixture 1:1, as a solvent for the binding material with 0.9 mg of o-NPOE, 2.5 mg polyvinyl chloride then 1.5 mg of the carbon powder, (5.0-15 mg) β -cyclodextrine (β -CD) and multiwall carbon nanotube (MWCT) ionophores added and after stirring for 15 min, the ink was sonicated and applied for printing of the electrodes. The influence of the plasticizer choice on the electrode performances has been studied as the electrode plasticized with o-NPOE is compared with those plasticized with DBP, DOP, DOS and TCP. The MSPEs were stored in a dry state at room temperature [31, 32].

Calibration of sensors

MCPEs and MSPEs were calibrated using standard BTFHC solutions with concentrations ranged from 1×10^{-7} to 1×10^{-2} mol L⁻¹ in a 25-mL beaker containing 2.0 mL acetate buffer solution of pH 5 at 25 °C with continuous stirring and the potential was recorded after stabilization to ± 0.1 mV and emf plotted as a function of logarithm BTFHC concentration. The graph was used for the subsequent determination of unknown concentration of BTFHC. The lower detection limit was taken at the point of intersection of the extrapolated linear segments of the BTFHC calibration curve.

Analytical method for pharmaceutical formulation

BTFHC cream was dissolved by minimum volume of warmed bidistilled water. The solution mixture was shaken in mechanical shaker and then transferred accurately to 50 mL measuring flask and completed to the mark with bidistilled water, shaken and finally determined by the proposed sensors. Then the content was estimated via potentiometric titration with NaTPB using MSPEs and MCPEs as sensing electrodes. The method was repeated several times to check the accuracy and reproducibility of the proposed method [33, 34].

Results and Discussion

Optimization of the electrode performance under batch conditions

The goal of this article is to fabricate novel MCPEs and MSPEs for the determination of BTFHC in pure form, pharmaceutical preparation and biological fluids. These electrodes have several advantages of low cost, fast response, low detection limit, high accuracy, wide concentration range, simple technique, quick preparation process which provide the possibility of measurements on small volumes as well as the ability for construction of a portable titration system, short response time and low Ohmic resistance. The sensitivity, linearity and selectivity of the ion selective electrodes (ISEs) obtained for a given paste depend significantly on nature and amount of plasticizers, ingredients of paste and amount of CNTs used. The effect of these parameters on behavior of the fabricated electrodes was studied and the results were provided in Table 1. The data obtained indicated that the developed electrodes can be successfully applied for the potentiometric determination of BTFHC in the concentration range from 1.57×10^{-7} to 1.0×10^{-2} mol L⁻¹ for MCPEs and 1.0×10^{-7} to $1.0 \times$

10^{-2} mol L⁻¹ for MSPEs, with Nernstian cationic slopes depending on the type of the electrode and method of fabrication. The limit of detection was found to be 1.5×10^{-7} for MCPEs (Sensors A and B) and 1.0×10^{-7} mol L⁻¹ for MSPEs (Sensors C and D). BTFH⁺ concentration with a slope of 56.95 ± 0.56 and 58.22 ± 0.78 mV decade⁻¹ for MCPEs (Sensors A and B) and 56.18 ± 0.66 and 57.69 ± 0.39 mV decade⁻¹ for MSPEs (Sensors C and D), respectively (Figure 1).

Effect of electrode composition

Optimization of BTFHC-MCPEs and BTFHC-MSPEs was performed by the selection of an optimum ratio of β -cyclodextrine (β -CD) ionophore. For this purpose, five electrodes were prepared containing different amounts of β -CD ionophore (5, 7.5, 10, 12.5 and 15 mg) which is the most important sensing component in an ion selective electrode; as it selectively binds the target ion while discriminating against interfering ions. The presence of multiwall carbon nanotubes in the composition of the MCPEs and MSPEs is to improve the conductivity of the sensor and to increase transduction of the chemical signal to electrical signal which in turn improve the dynamic working range and response time of the sensors to Nernstian slope values. These results

indicated that the highest potential break at the end point was evaluated using 12.5 mg of β -CD ionophore for MCPE (electrode A) and MSPE (electrode C) sensors, respectively. But when increasing the amount of ionophore over 12.5 mg, the total potential change decreased as shown in Figure 2.

Effect of soaking

Electrode must be soaked in order to activate the surface of the MCPE and MSPE layers to form an infinitesimally thin gel layer at which ion exchange occurs. The MCPEs and MSPEs were soaked in ion-pair suspended solution, and the titration curves were plotted from which the total potential changes were recorded after 0 (without soaking), 5, 15, 30 min, 2 h and 24 h. Studying the effect of soaking time on the BTFHC electrodes showed that the optimum soaking time is 15 min for all investigated electrodes (Figure 3). Soaking for longer time than 15 min was not recommended in order to avoid leaching of the very little amount of the electroactive species into the bathing solution. The electrodes were kept dry in an opaque closed vessel and stored in a refrigerator while not in use. The reproducibility of repeated measurements on the same solutions was ± 1 mV.

TABLE 1: Response characteristics of BTFHC-MCPEs (electrodes A and B) and BTFHC-MSPEs (electrodes C and D).

Parameter	BTFHC- MCPE		BTFHC-MSPE	
	Electrode A	Electrode B	Electrode C	Electrode D
Slope (mV decade ⁻¹)	56.95±0.56	58.22±0.78	56.18±0.66	57.69±0.39
Usable range (mol L ⁻¹)	$1.57 \times 10^{-7} - 1.0 \times 10^{-2}$		$1.0 \times 10^{-7} - 1.0 \times 10^{-2}$	
Detection limit (mol L ⁻¹)	1.57×10^{-7}		1.0×10^{-7}	
Response time (s)	12		7	
Working pH range	5.0 - 9.0		4.0 - 9.0	
SD of slope (mV decade ⁻¹)	0.302	0.174	0.351	0.209
Intercept (mV)	454.64±1.19	493.44±1.11	497.92±1.17	538.61±0.87
Life time (days)	82	99	160	185
Accuracy (%)	99.95	99.89	99.93	99.97
Precision (%)	0.173	0.161	0.193	0.177

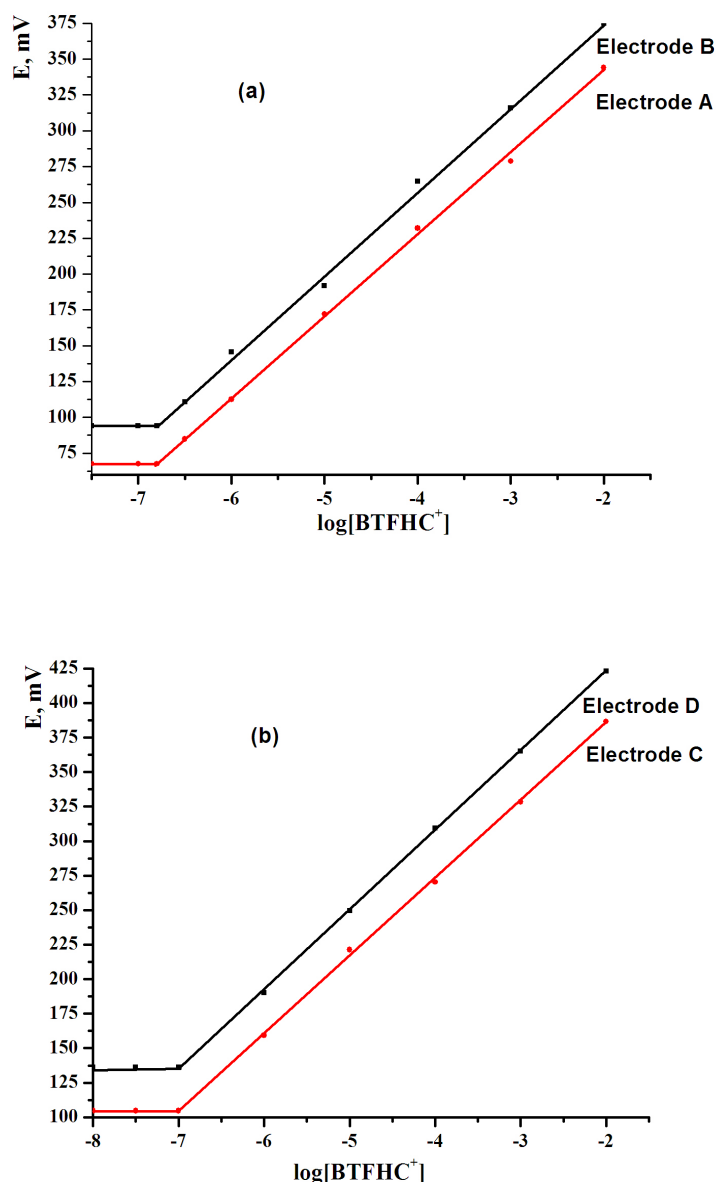


Fig.1: Calibration graphs using (a) MCPEs (electrodes A and B) and (b) MSPEs (electrodes C and D).

Effect of plasticizer type

The plasticizer is an important constituent; therefore a study of influence of plasticizers on the potentiometric response characteristics of the proposed electrodes was investigated. Five plasticizers with different polarities namely o-NPOE, TCP, DBP, DOP and DOS were used. The results in Figure 4 showed that, from the all tested plasticizers, o-NPOE and TCP exhibited the largest potential change, the highest potential break at the end point of the electrodes and the shortest response time in comparison with other plasticizers. This may be attributed to o-NPOE and

TCP have a greater polarity and less lipophilicity. This pointed out that the presence of plasticizers improved the workability of the electrodes. Electrodes plasticized with DBP, DOP and DOS showed poor sensitivities which can accounted to low solubility or low distributions of ion-pairs in them.

Effect of pH

The effect of pH on the performance of the fabricated electrodes at concentration of 1×10^{-3} and 1×10^{-5} mol L^{-1} BTFHC solution was investigated (Figure 5). The operational

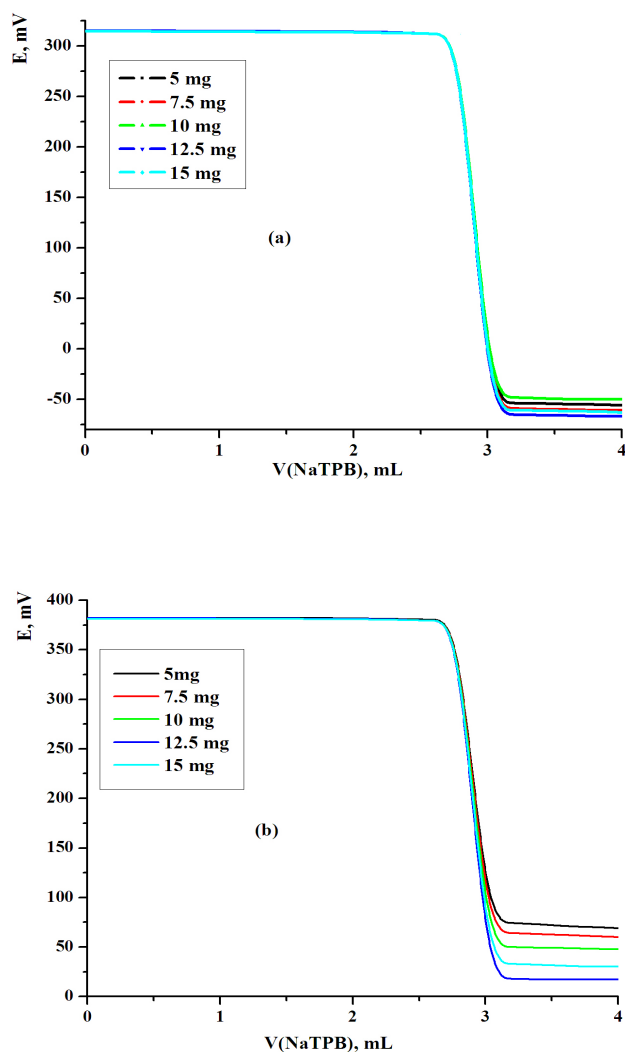


Fig. 2: Effect of ionophore contents on (a) Electrode A and (b) Electrode C using TCP plasticizer.

range was studied by adjusting the pH of drug solution with dilute HCl and/or NaOH ($0.1\text{--}1.0\text{ mol L}^{-1}$ each). The potentials remained constant in pH range of $5.0\text{--}9.0$ for sensors (A and B) and in pH range of $4.0\text{--}9.0$ for sensors (C and D). In this range the electrode can be safely used for BTFH⁺ determination by using buffer solutions of the suitable pH within this working range. No significant change in the potential pH behavior is observed during the working period of the electrodes. At lower pH values at $\text{pH} < 4$, the decrease in mV readings may be due to the interference of hydronium ion. While, at higher pH values at $\text{pH} > 9.0$, the decrease in the mV readings was due to formation of non-protonated drug molecules and the hydroxyl of the BTFH⁺

is formed in the test solution, so lower e.m.f. readings were recorded (Figure 5).

Effect of temperature on the electrode performance

The thermal stability of the electrodes was studied by demonstrating the effect of temperature (from 10 to $60\text{ }^{\circ}\text{C}$) on their performance. The electrode potentials of for, were determined. Calibration graphs (electrode potential (E_{cell}) versus $\text{p}[\text{BTFHC}]$) were constructed for 1.57×10^{-7} – $1.0 \times 10^{-2}\text{ mol L}^{-1}$ and 1.0×10^{-7} – $1.0 \times 10^{-2}\text{ mol L}^{-1}$ of BTFHC test solutions using MCPes and MSPes, respectively. For the determination of the isothermal coefficient (dE°/dT) of the electrode, the standard electrode potential (E_{elec}°) at different temperatures was plotted versus ($t-25$), where this is the temperature of the test solution. A straight-

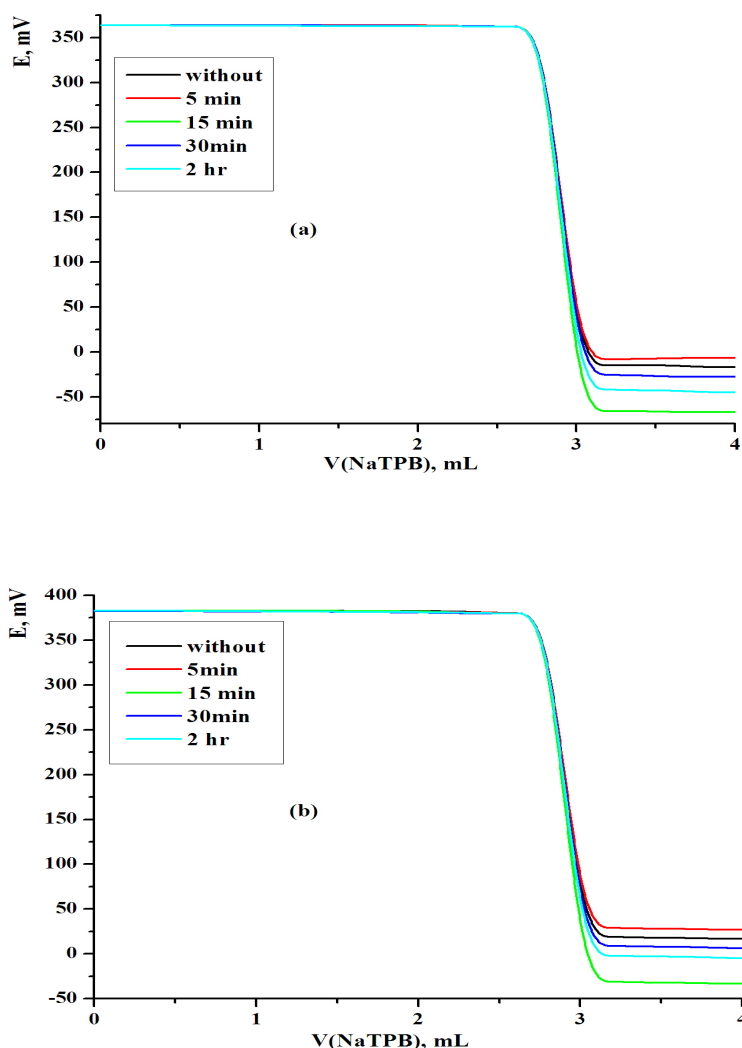


Fig. 3: Effect of soaking time on (a) Electrode A and (b) Electrode C using TCP plasticizer.

line plot was obtained according to the equation (35)

$$E^{\circ} = E^{\circ}_{(25)} + (dE^{\circ}/dt)(t - 25)$$

Where $E^{\circ}_{(25)}$ is the standard electrode potential at 25 °C, the slope of the straight-line obtained represents the isothermal coefficient of the electrodes which were found to be 0.000257, 0.000228, 0.000199 and 0.000121 mV/°C for electrodes A, B, C and D, respectively, (Figure 6). The small values of the isothermal coefficient of the electrodes indicated the high thermal stability of the electrodes within the investigated temperature range as they were found to be usable up to 60 °C without noticeable deviation from the Nernstian behavior.

Response time

The static response time of the electrodes was measured after successive immersions in a series of solutions, each having a 10 fold concentration difference in order to achieve a steady potential within ± 1 mV of the final steady-state value. The dynamic response time of the electrodes (A-D) was determined for the concentration range 1.0×10^{-6} - 1.0×10^{-3} mol L⁻¹ for MCPes and MSPes (Figure 7). The response time was found to be 12 s for MCPes (electrodes A and B) and 7 s for MSPes (electrodes C and D), respectively.

Lifetime and reproducibility

From the data obtained during the long time period of 82, 99, 160 and 185 days (Figure 8) for electrodes A, B, C and D, respectively, the

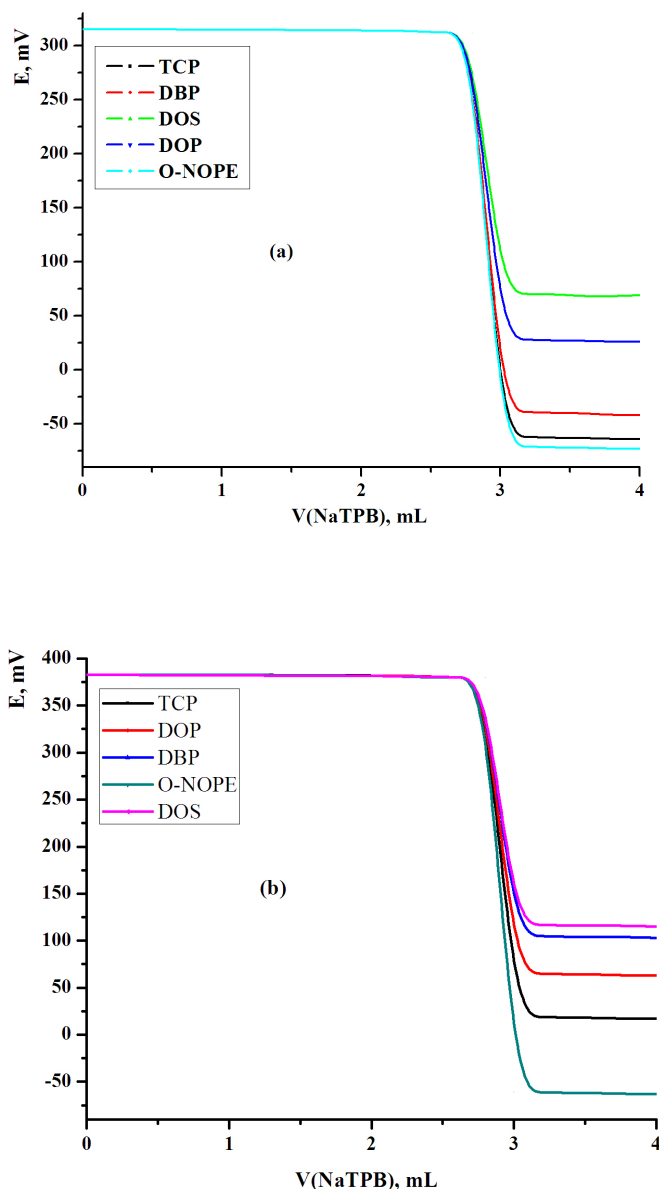


Fig. 4: Effect of plasticizer type on (a) Electrode A and (b) Electrode C.

slope of the sensors after this period drifted away from Nernstian behavior. This may be attributed to the decrease in the quantity of ionophore and the plasticizer in the paste due to the migration of these components. It was pointed out from these results that BTFHC-MCPEs and MSPEs can be used over a period of three and six months without any change in the value of the slope, working concentration range and detection limit.

Selectivity coefficient of the electrodes

Selectivity is the most important characteristic of ISEs which described their specificity toward

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the target ion in the presence of interfering ions. The potentiometric selectivity coefficient of an electrode is defined by its relative response for the primary ions over other ions present in the solution. The selectivity coefficients for BTFHC with regard to a variety of some inorganic cations, sugar species, organic and glycine under the optimized experimental conditions, were investigated (Table 2). The selectivity coefficients of the proposed modified electrodes were measured using the separate solutions (SSM) and matched potential (MPM) methods with 1.0×10^{-3}

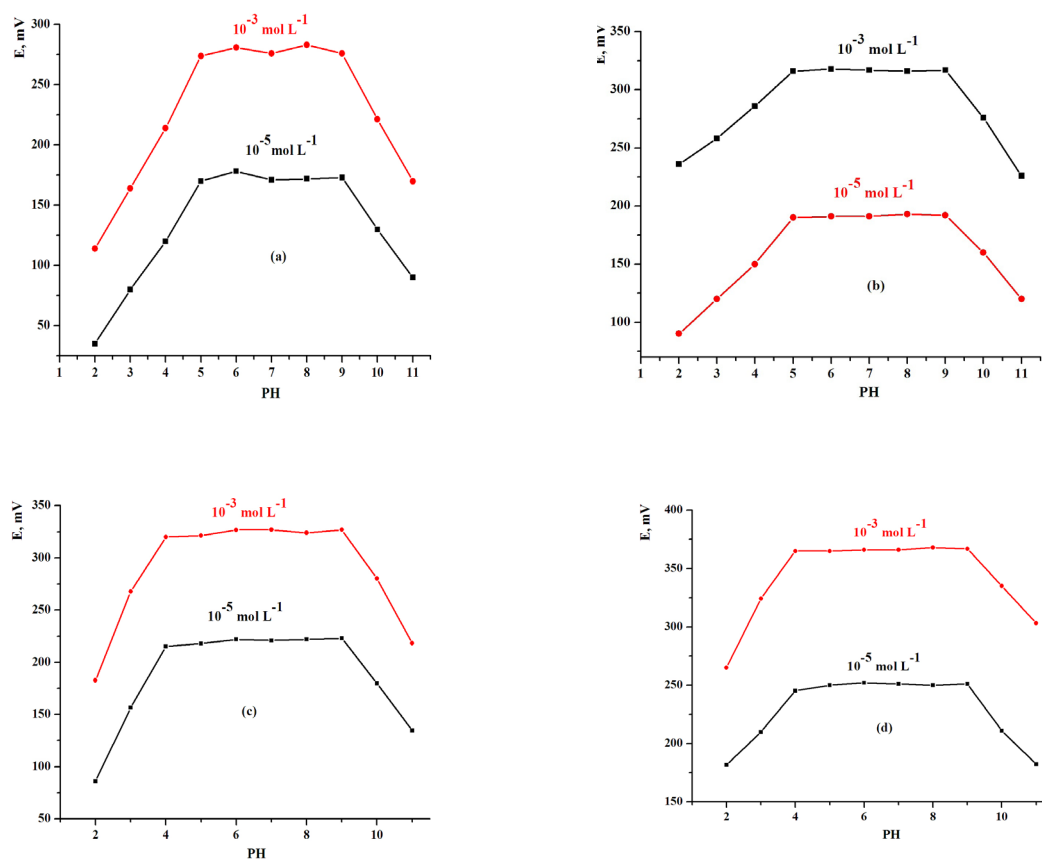


Fig.5: Effect of pH of the test solution on the performance of MCPEs [(a) electrode (A) and (b) electrode (B)] and MSPEs [(c) electrode (C) and (d) electrode (D)].

TABLE 2: Potentiometric selectivity coefficients of some interfering ions using MCPEs (electrodes A and B) and MSPEs [electrodes C and D].

	MCPEs		Interfering ions		$-\log K_{\text{BTFHC}, \text{B}}^{\text{SSM}}$	Interfering ions		$-\log K_{\text{BTFHC}, \text{B}}^{\text{MPM}}$	
	Electrode (A)	Electrode (B)	Electrode (C)	Electrode (D)		Electrode (A)	Electrode (B)	Electrode (C)	Electrode (D)
Na^+	3.78	3.89	3.72	3.86	Sucrose	2.75	2.81	2.53	2.79
K^+	3.56	3.66	3.53	3.65	Maltose	2.93	3.01	2.87	3.05
Ca^{2+}	3.03	3.22	3.05	3.13	Glycine	2.18	2.36	2.21	2.42
Zn^{2+}	4.06	4.15	3.99	4.12	Picric acid	3.22	3.45	3.14	3.42
Cu^{2+}	2.78	2.96	2.63	2.93	Glucose	3.60	3.77	3.59	3.74
Co^{2+}	3.77	3.86	3.79	3.83	Fructose	2.99	3.07	3.02	3.06
Ni^{2+}	3.34	3.46	3.31	3.42	Lactose	2.61	2.69	2.57	2.64
Cd^{2+}	3.92	3.98	3.87	3.85	Urea	2.99	3.09	3.00	3.12
Mn^{2+}	4.11	4.27	4.06	4.22	Starch	3.57	3.66	3.48	3.62
Fe^{3+}	2.08	2.13	2.10	2.22					
Al^{3+}	3.09	3.33	3.23	3.41					

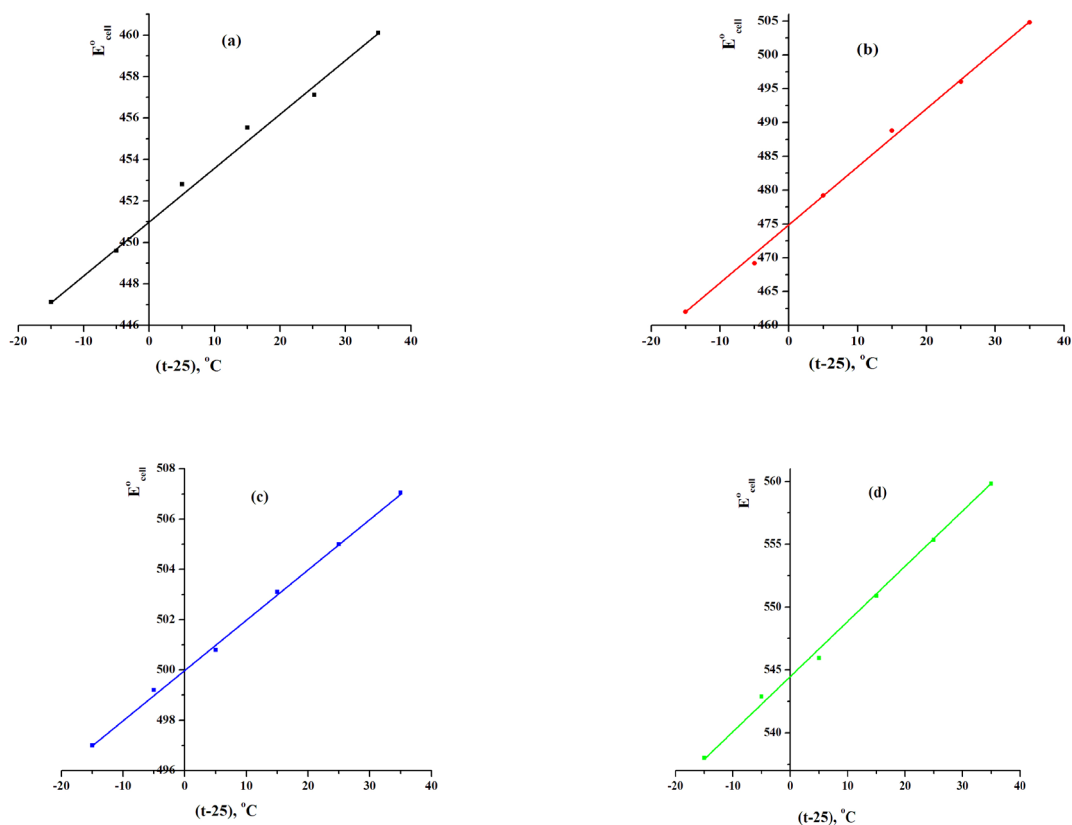


Fig.6: Effect of temperature on the performance of MCPEs [(a) electrode (A) and (b) electrode (B)] and MSPEs [(c) electrode (C) and (d) electrode (D)].

concentration of both the standard drug and the interference. The selectivity coefficient, $\log K_{\text{BTFH}^+, \text{B}}^{\text{pot}}$, of the MCPEs and MSPEs are determined by employing separate solution method (SSM) with the rearranged Nicolsky equation [36]:

$$\log K_{\text{BTFH}^+, \text{B}}^{\text{pot}} = (E_2 - E_1) / S + (1 + z_1 / z_2) \log a$$

Where, E_1 is the potential measured in $1.0 \times 10^{-3} \text{ mol L}^{-1}$ BTFH⁺ cation, E_2 the potential measured in $1.0 \times 10^{-3} \text{ mol L}^{-1}$ of the interfering compound (B), z_1 and z_2 are the charges of the BTFH⁺ cation and interfering species (B), respectively, S is slope of the electrode calibration plot, and a is the concentration of the ions used ($1 \times 10^{-3} \text{ mol L}^{-1}$). Also, the selectivity of the investigated electrodes was determined by the matched potential method (MPM). In this method, the potentiometric selectivity coefficient is defined as the activity ratio of primary and interfering ions (BTFH⁺) and (B) that give the same potential change under identical conditions. The results obtained (Table 2) showed high selectivity, high sensitivity and showed no significant interference from glycine,

maltose, picric acid, urea, starch, sucrose, fructose and lactose. The inorganic cations did not interfere due to the differences in their ionic size and hence their mobilities, polarities and permeabilities as compared to those of BTFH⁺ cation.

Analytical applications to drug analysis

To test the analytical applicability of the proposed sensors, it has been applied for three different pharmaceutical samples of BTFHC drug (Tinearest, Bravamax and Farbicure) by using the potentiometric titration and standard addition methods. The results obtained were compared to the British Pharmacopiea and the data obtained showed low values of SD%, RSD% and gives high precision and accuracy of the proposed potentiometric method (Table 3). F-test was used for comparing precision of the two methods and t-test was used for comparing the accuracy.

Application to serum and urine

The proposed potentiometric method was applied to determine BTFHC in biological fluids such as human serum and urine (Table 4). The

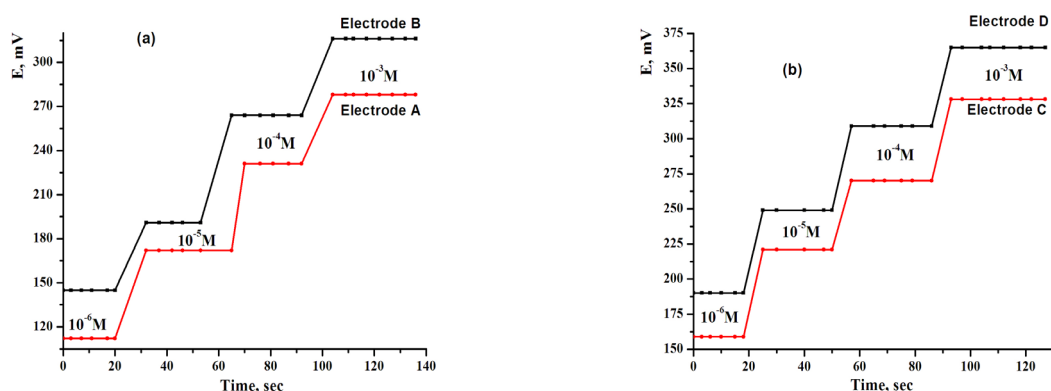


Fig.7: Dynamic response time of BTFHC electrodes [(a) electrode (A) and electrode (B)] and (b) electrode (C) and electrode (D)].

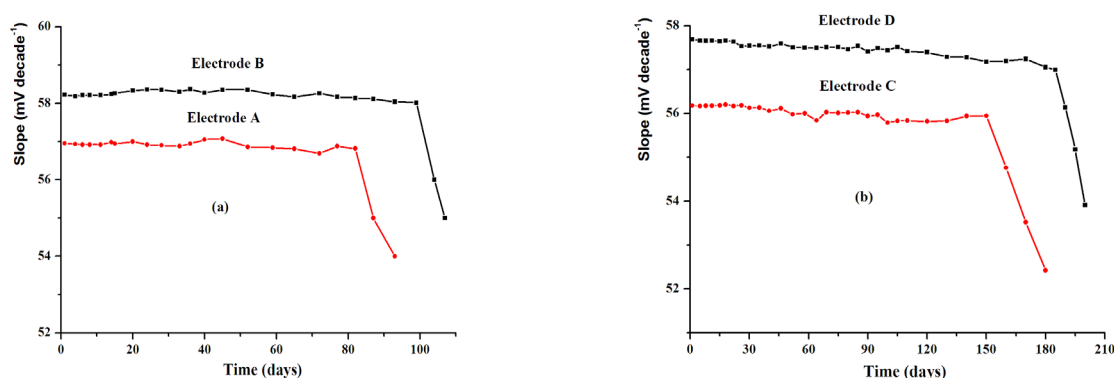


Fig.8: Effect of life time on the performance of BTFHC electrodes [(a) electrode (A) and electrode (B)] and (b) electrode (C) and electrode (D)].

TABLE 3. Potentiometric determination of BTFHC in pharmaceutical formulations using MCPs (electrodes A and B) and MSPEs (electrodes C and D).

Sample No.	[BTFHC] mg/mL Pharmaceutical Preparation					RSD(%)				
	British Pharmacopeia	A	B	C	D	British Pharmacopeia	A	B	C	D
1	0.452	0.451	0.455	0.449	0.453	0.731	0.756	0.538	0.804	0.559
2	0.481	0.483	0.485	0.482	0.486	0.806	0.674	0.621	0.692	0.633
3	0.424	0.426	0.428	0.425	0.427	0.829	0.668	0.616	0.670	0.622

SD values for Pharmaceutical Preparation (British Pharmacopeia = 0.089-0.235), (electrode A = 0.091-0.248) (electrode B = 0.035-0.189), (electrode C = 0.097- 0.302) and (electrode D = 0.038- 0.196).

F-test = (electrode A = 0.9 – 1.3), (electrode B = 0.1 – 0.7), (electrode C = 0.4 – 1.1) and (electrode D = 0.2 – 0.9). (Tabulated F value at 95% confidence limit = 3.52 for n = 4).

t-test = (electrode A = 1.0 – 1.8), (electrode B = 0.2 – 0.8), (electrode C = 1.1 – 1.7) and (electrode D = 0.3 – 1.2). (Tabulated t value at 95% confidence limit = 1.978 for n = 4).

TABLE 4: Determination of BTFHC in spiked urine and human serum using MCPE (Electrode B) and MSPE (Electrode D)

Sample	Statistical parameters	(Electrode B)			(Electrode D)		
		Direct method	Calibration graphs	Standard addition method	Direct method	Calibration graphs	Standard addition method
urine	Mean recovery (%)	99.64	99.35	99.05	99.41	99.11	98.98
	N	5	5	5	5	5	5
	Variance	0.53	0.49	0.61	0.55	0.46	0.63
	RSD (%)	0.17	0.26	0.33	0.26	0.37	0.49
serum	Mean recovery (%)	99.80	99.66	99.23	99.77	99.52	99.18
	N	5	5	5	5	5	5
	Variance	0.27	0.35	0.46	0.39	0.42	0.49
	RSD (%)	0.12	0.23	0.29	0.33	0.36	0.39

accuracy of the proposed sensors (B and D) was investigated by the determination of BTFHC in spiked urine and serum samples prepared from serial concentrations of BTFHC reference standards. The results showed that the proposed potentiometric method is accurate and precision for the determination of BTFHC in urine and serum samples without interferences caused by the excipients expected to be present in drug as indicated by the percentage recovery values.

Precision and accuracy

In order to determine the precision of the proposed method, solutions containing concentrations of BTFHC were prepared and analyzed and the analytical results are summarized in Table 1. The results showed high accuracy of values 99.95, 99.89, 99.93 and 99.97% and good precision values of 0.173, 0.161, 0.193 and 0.177 % for electrodes A, B, C and D, respectively, and low values of RSD% and SD values were obtained within the same day to evaluate repeatability (intra-day precision) and over five days to evaluate intermediate precision (inter-day precision).

References

1. Porras, A. M. G., Terra, B. S., Braga, T. C., Magalhães, T. F. F., Martins, C. V. B., da Silva, D. L., Baltazar, L. M., Gouveia, L. F., de Freitas, G. J. C., Santos, D. A., and other authors: Butenafine and analogues: An expeditious synthesis and cytotoxicity and antifungal activities, *Journal of Advanced Research*, 14, 81-91 (2018).
2. Miron, D., Cornelio, R., Troleis, J., Mariath, J., Zimmer, A. R., Mayorga, P., and Schapoval, E. S.: Influence of penetration enhancers and molecular weight in antifungals permeation through bovine hoof membranes and prediction of efficacy in human nails, *European Journal of Pharmaceutical Sciences*, 51, 20-25 (2014).
3. Pillai, A. B., Nair, J. V., Gupta, N. K., and Gupta, S.: Microemulsion-loaded hydrogel formulation of butenafine hydrochloride for improved topical delivery, *Archives of Dermatological Research*, 307, 625-633 (2015).
4. Beydoun, K., Ghattas, G., Thenert, K., Klankermayer, J., and Leitner, W.: Ruthenium-catalyzed reductive methylation of imines using carbon dioxide and molecular hydrogen, *Angewandte Chemie-International Edition*, 53, 11010-11014 (2014).
5. Mitra, A., Kim, N., Spark, D., Toner, F., Craig, S., Roper, C., and Meyer, T. A.: Use of an in vitro human skin permeation assay to assess bioequivalence of two topical cream formulations containing butenafine hydrochloride (1%, w/w), *Regulatory Toxicology and Pharmacology*, 82, 14-19 (2016).

6. Robert, M. E. M. and Kalia, Y. N.: New developments in topical antifungal therapy, *American Journal of Drug Delivery*, 4, 231-247 (2006).
7. Heikal, E. K., Elmelawy, M. S., Khalil, S. A., and Elbasuny, N. M.: Manufacturing of environment friendly biolubricants from vegetable oils, *Egyptian Journal of Petroleum*, 26, 53-59 (2017).
8. Rao, S., Barot, T., Rajesh, K. S., and Jha, L. L.: Formulation, optimization and evaluation of microemulsion based gel of Butenafine Hydrochloride for topical delivery by using simplex lattice mixture design, *Journal of Pharmaceutical Investigation*, 46, 1-12 (2016).
9. Ali, T. A., Mohamed, G. G., and Yahya, G. A.: Development of novel potentiometric sensors for determination of lidocaine hydrochloride in pharmaceutical preparations, serum and urine samples, *Iranian Journal of Pharmaceutical Research*, 16, 498-512 (2017).
10. Frag, E. Y. Z., Ali, T. A., Mohamed, G. G., and Awad, Y. H. H.: Construction of different types of ion-selective electrodes. characteristic performances and validation for direct potentiometric determination of orphenadrine citrate, *International Journal of Electrochemical Science*, 7, 4443-4464 (2012).
11. Ali, T. A., Mohamed, G. G., Eldessouky, H., and Adeeb, A. E.: Ion selective electrodes based on methyl 6-(hydroxymethyl) picolinate ionophore for electrochemical determination of Fe(III) in petroleum water samples, *Egyptian Journal of Petroleum*, 28, 233-239 (2019).
12. Bangaleh, Z., Sadeghi, H. B., Ebrahimi, S. A., and Najafizadeh, P.: A new potentiometric sensor for determination and screening phenylalanine in blood serum based on molecularly imprinted polymer, *Iranian Journal of Pharmaceutical Research*, 18, 61-71 (2019).
13. Cavus, I. and Kanberoglu, G. S.: Development of a potentiometric maprotiline-selective electrode and its application in pharmaceutical samples, *Microchemical Journal*, 148, 57-65 (2019).
14. Ali, T. A., Mohamed, G. G., Al-Sabagh, A. M., and Migahed, M. A.: A new screen-printed ion selective electrode for determination of citalopram hydrobromide in pharmaceutical formulation, Fenxi Huaxue/ *Chinese Journal of Analytical Chemistry*, 42, 565-572 (2014).
15. Ali, T. A., Mohamed, G. G., Omar, M. M., and Abdrabou, V. N.: Improved determination of mebeverine hydrochloride in urine, serum and pharmaceutical preparations utilizing a modified carbon paste electrode, *International Journal of Electrochemical Science*, 10, 2439-2454 (2015).
16. Abu Shawish, H. M., Tamous, H., Saadeh, S. M., and Tbaza, A.: Determination of atomoxetine hydrochloride in biological fluids using potentiometric carbon paste electrode modified by TiO₂ nanoparticles, *Acta Chimica Slovenica*, 65, 811-822 (2018).
17. Khamees, N., Mohamed, T. A. F., Derar, A. R., and Aziz, A.: All-solid-state, PVC membrane, and carbon paste ion-selective electrodes for determination of donepezil hydrochloride in pharmaceutical formulation, *Journal of AOAC International*, 100, 1414-1419 (2017).
18. Rouhani, M. and Soleymanpour, A.: A new selective carbon paste electrode for potentiometric analysis of olanzapine, Measurement: *Journal of the International Measurement Confederation*, 140, 472-478 (2019).
19. Shabani, R., Rizi, Z. L., and Moosavi, R.: Selective potentiometric sensor for isoniazid ultra-trace determination based on Fe₃O₄ nanoparticles modified carbon paste electrode (Fe₃O₄/CPE), *International Journal of Nanoscience and Nanotechnology*, 14, 241-249 (2018).
20. Akl, Z.F. and Ali, T.A.: A novel modified screen-printed electrode with triazole surfactant assembled on silver nanoparticles for potentiometric determination of uranium, *Journal of Radioanalytical and Nuclear Chemistry*, 1-11 (2017).
21. Ali, T. A., Aglan, R. F., Mohamed, G. G., and Mourad, M. A.: New Chemically modified screen-printed electrode for co(II) determination in different water samples, *International Journal of Electrochemical Science*, 9, 1812-1826 (2014).
22. Ali, T. A. and Mohamed, G. G.: Modified screen-printed ion selective electrodes for potentiometric determination of sodium dodecylsulfate in different samples, *Journal of AOAC International*, 98, 116-123 (2015).
23. Mohamed, G. G., Ali, T. A., El-Shahat, M. F., Al-Sabagh, A. M., Migahed, M. A., and Khaled, E.: Potentiometric determination of cetylpyridinium chloride using a new type of screen-printed ion selective electrodes, *Analytica Chimica Acta*, 673, 79-87 (2010).

24. Mohamed, G. G., Ali, T. A., El-Shahat, M. F., Migahed, M. A., and Al-Sabagh, A. M.: Novel screen-printed electrode for the determination of dodecyltrimethylammonium bromide in water samples, *Drug Testing and Analysis*, 4, 1009-1013 (2012).
25. Akl, Z. F. and Ali, T. A.: Highly sensitive potentiometric sensors for thorium ions detection using morpholine derivative self-assembled on silver nanoparticles, *RSC Advances*, 6, 77854-77862 (2016).
26. Ali, T. A., Azzam, E. M. S., Hegazy, M. A., El-Faragy, A. F. M., and Abd-elaal, A. A.: Zinc(II) modified carbon paste electrodes based on self-assembled mercapto compounds-gold-nanoparticles for its determination in water samples, *Journal of Industrial and Engineering Chemistry*, 20, 3320-3328 (2014).
27. Ali, T. A., Soliman, M. H., Mohamed, G. G., Farag, A. B., and Samah, M. K.: Development of a new modified screen-printed and carbon paste electrodes for selective determination of cetyltrimethylammonium bromide in different water samples, *International Journal of Electrochemical Science*, 10, 3192-3206 (2015).
28. Khaled, E., Mohamed, G. G., and Awad, T.: Disposal screen-printed carbon paste electrodes for the potentiometric titration of surfactants, *Sensors and Actuators, B: Chemical*, 135, 74-80 (2008).
29. Mohamed, G. G., Ali, T. A., El-Shahat, M. F., Al-Sabagh, A. M., and Migahed, M. A.: New Screen-Printed Ion-Selective Electrodes for Potentiometric Titration of Cetyltrimethylammonium Bromide in Different Civilic Media, *Electroanalysis*, 22, 2587-2599 (2010).
30. Mohamed, G. G., El-Shahat, M. F., Al-Sabagh, A. M., Migahed, M. A., and Ali, T. A.: Septonex-tetraphenylborate screen-printed ion selective electrode for the potentiometric determination of Septonex in pharmaceutical preparations, *Analyst*, 136, 1488-1495 (2011).
31. Ali, T. A., Hassan, A. M. E., and Mohamed, G. G.: Manufacture of lead-specific screen-printed sensor based on lead schiff base complex as carrier and multi-walled carbon nanotubes for detection of Pb(II) in contaminated water tests, *International Journal of Electrochemical Science*, 11, 10732-10747 (2016).
32. Ali, T. A., Mohamed, G. G., and Said, A. H.: *Egypt. J. Chem.* 63, No. 8 (2020)
33. Shaikshavali, P., Madhusudana Reddy, T., Venu Gopal, T., Venkataprasad, G., Kotakadi, V. S., Palakollu, V. N., and Karpoormath, R.: A simple sonochemical assisted synthesis of nanocomposite (ZnO/MWCNTs) for electrochemical sensing of Epinephrine in human serum and pharmaceutical formulation, *Colloids and Surfaces A: Physicochemical and Engineering Aspects*, 584, 124038 (2020).
34. Thapliyal, N. B., Chiwunze, T. E., Karpoormath, R., and Cherukupalli, S.: Fabrication of highly sensitive gold nanourchins based electrochemical sensor for nanomolar determination of primaquine, *Materials Science and Engineering: C*, 74, 27-35 (2017).
35. Antropov, L. I. and Beknazarov, A.: Theoretical electrochemistry, in: University Press of the Pacific, Honolulu (2001).
36. Bakker, E., Pretsch, E., and Bühlmann, P.: Selectivity of Potentiometric Ion Sensors, *Analytical Chemistry*, 72, 1127-1133 (2000).