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RESEARCH ARTICLE

Development of a New Coated Graphite Electrode for Hydroxychloroquine Sulfate Determination in Pharmaceutical Preparations and Human Urine

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Abstract

A simple, rapid and sensitive method for the determination of hydroxychloroquine sulfate using coated graphite electrode (CGE) was developed. The membrane film of the electrode made of 5.0% (w/w) ion-pair of hydroxychloroquine sulfate (HCQS) with sodium tetraphenylborate (NaTPB) and 47.5% (w/w) of each of dibutyl phthalate (DBP) as plasticizing solvent and poly vinyl chloride (PVC). The electrode showed Nernstian slope of 30.0±0.2 mV decade^{-1} at 25±0.1°C within the concentration range of 7.1×10^{-5} to 1.0×10^{-2} mol L^{-1} (HCQS), with a detection limit of 1.8×10^{-5} mol L^{-1} HCQS and response time ≤ 10 s. Up to 24 h continuous soaking, the calibration graph slope was constant at 30.0±0.2 mV decade^{-1} at 25±0.1°C, then it decreased gradually as reaching 23.6±0.6 mV decade^{-1} after 20 days. The changes in pH did not affect the electrode performance within the range of 2-7. The standard electrode potentials were determined at different temperatures and used to calculate the thermal coefficient of the electrode. The electrode showed very good selectivity for HCQ cation with respect to a number of common inorganic and organic species. The standard addition method and potentiometric titration were used to determine HCQS in pure solutions, pharmaceutical formulations and human urine.

INTRODUCTION

Hydroxychloroquine sulfate (HCQS) (Fig. 1), 2-((4-((7-chloro-4-quinolinyl) amino)pentyl)ethylamino)ethanol sulfate(1:1),is an antimalarial drug indicated for the suppressive treatment and the treatment of acute attacks of malaria due to *P. vivax*, *P. malariae*, *P. ovale*, and susceptible strains of *P. falciparum* [1,2]. It is also indicated for the treatment of rheumatoid arthritis [3, 4] and discoid and systemic lupus erythematosus [5]. Determination of hydroxychloroquine sulfate includes extraction followed by nonaqueous titrimetric [6] or spectrophotometric methods based on reactions with iodide [7], quinones [8, 9] and cobalt thiocyanate [10]. Because of the nonspecificity of most of these reactions, prior extraction of HCQ is commonly involved in the assay methods. Besides, a number of different HPLC methods have been proposed for the determination of HCQ [11] in biological fluids [12] using spectrophotometry [13], fluorescence [14, 15, 16], ultraviolet [17] or tandem mass spectrometry [18] detection. HCQ was detected also using a differential pulse voltammetry method [19]. Few electrochemical methods have been used for determination of aminoquinolines. Among them can be mentioned the potentiometric determination of the chloroquine (CQ) in various pharmaceutical preparations using an ion-selective membrane electrode [20]. A recent review of the literature found no reports on potentiometric determination of hydroxychloroquine sulfate.

Ion-selective electrodes (ISEs) have found wide spread use for the direct determination of ionic species [21-31]. Coated electrodes in which an electroactive species is incorporated into a thin polymeric film coated directly on a metallic or graphite conductor have been shown to be very effective for a wide variety of inorganic and organic ions [32-37]. Electrodes of this sort are simple, inexpensive, durable, capable of reliable response in a wide concentration range for a variety of both organic and inorganic ions and suitable for measurements in small volumes of sample or for the desired in vivo applications of ISEs that biomedical researchers have long awaited.

The present work describes construction and investigation of performance characteristics of new coated graphite electrode (CGE) for the determination of hydroxychloroquine sulfate in pure form, pharmaceutical preparations and human urine samples.

2. Experimental

2.1. Reagents and Materials

All the chemicals used were of analytical grade. Bidistilled water was used throughout all experiments. Dibutyl phthalate (DBP) was purchased from Merck. Sodium tetraphenylborate (NaTPB) was obtained from Fluka. Poly vinyl chloride (PVC) of high molecular weight and tetrahydrofuran (THF) were purchased from Aldrich Chemical Company (USA). The metal salts were provided by BDH Company (UK) as nitrates or chlorides. Stock solutions of the metal salts were prepared in bidistilled water and standardized whenever necessary.

Pure-grade hydroxychloroquine sulfate (M. wt = 433.95 g mol^{-1}) was supplied by MINAPHARM Co., Cairo, Egypt. The pharmaceutical preparation (Hydroquine 200 mg/tablet) was purchased from local drug stores. Standard solution of 1.0×10^{-2} mol L^{-1} hydroxychloroquine sulfate was freshly prepared by dissolving the accurately weighed amount in bidistilled water. Working solutions of the drug (1.0×10^{-6} – 1.0×10^{-2} mol L^{-1}) were prepared by suitable dilution from the standard solution with bidistilled water.

Stock solution of 1.0×10^{-2} mol L^{-1} NaTPB was prepared by dissolving the accurately weighed amount of the pure solid in bidistilled water. Solutions of sodium hydroxide and hydrochloric acid of concentrations within the range (0.1-1.0) mol L^{-1} were used for adjusting the pH of the medium.

2. 2. Apparatus

The electrochemical system of the CGE may be represented as follows:

Graphite rod/membrane/test solution/ Ag/AgCl double-junction reference electrode

An Ag/AgCl double-junction reference electrode (Metrohm 6.0222.100) was used as the external reference. Potentiometric and pH-measurements were carried out using 702 titroprocessor equipped with a 665 dosimat made by Metrohm (Switzerland). A mLw W20 circulator thermostat was used to control the temperature of the test solutions.

2. 3. Preparation of the Ion-Pair Compound

The ion-pair compound, HCQ-TPB was prepared by slow addition of 50 mL of 1.0×10^{-2} mol L^{-1} sodium tetraphenylborate solution to 100 mL of 1.0×10^{-2} mol L^{-1} hydroxychloroquine sulfate under stirring for 15 min. The resulting precipitate was filtered off through a Whatman filter paper No. 42, washed with bidistilled water several times until sulfate free (tested using $BaCl_2$ solution) , dried at room temperature and ground to fine powder. The composition of the ion-pair was confirmed by elemental analysis to be 1:2 (HCQ:TPB).

2. 4. Conductometric Measurements

Conductometric titration was followed with a Jenway conductivity meter. 50 mL of 1.0×10^{-3} mol L^{-1} hydroxychloroquine sulfate solution was transferred to the 100-mL cell and the solution titrated against a 1.0×10^{-2} mol L^{-1} NaTPB solution using a microburette. The conductance of the solution was measured after each addition of the titrant. Conductance values were corrected by multiplying by the dilution coefficient and plotted versus molar ratio. The titration plot showed a break which corresponds to the stoichiometry of the ion-pair.

2. 5. Electrode Construction

CWEs were constructed using silver, copper and aluminium metal wires (1 mm diameter) and graphite rod (4 mm diameter) following the procedures described in details elsewhere [38]. One of the two ends of each rod was used for connection while the other, about 2 cm length, was dipped in the coating solution and left to dry in air. The process was repeated several times till a layer of the proper thickness was formed covering the terminal of the rod.

The prepared electrode was preconditioned by soaking for 30 min in 1.0×10^{-3} mol L^{-1} hydroxychloroquine sulfate solution. When not in use, the electrode was stored in air.

2. 6. Construction of Calibration Curves

The conditioned electrode was immersed in conjunction with an Ag/AgCl double-junction reference electrode in solutions of hydroxychloroquine sulfate in the range of 1.0×10^{-6} -1.0×10^{-2} mol L^{-1}. The electrode was allowed to equilibrate whilst stirring and recording the e.m.f. readings within ±1 mV. The mV- concentration profiles were plotted. The regression equations for the linear part of the curves were computed and used for subsequent determination of unknown concentrations of hydroxychloroquine sulfate.

2. 7. Selectivity

The potentiometric selectivity coefficients ($K^{pot}_{Prx,j}$) of hydroxychloroquine electrode were evaluated at different concentrations of both hydroxychloroquine and the interferents according to IUPAC recommendations using the separate solutions method (SSM) in case of inorganic cations (Li^+, K^+, Mg^{2+}, Ca^{2+}, Co^{2+}, Mn^{2+}, Fe^{2+}, Cu^{2+},NH_4^+) [39], and the matched potential method (MPM) [40] in case of neutral species (amino acids, sugars, vitamins, and urea). In the separate solution method, the Nicolsky Eisenman equation [41] was used:

$$K^{pot}_{Prx,j} = (E_2 - E_1)/S + \log [Prx] - \log [J^{z+}]^{1/z}$$

Where, E_1 and E_2 are the electrode potentials in a $1.0x10^{-3}$ mol L^{-1} solution of hydroxychloroquine sulfate and J^{Z+} interfering ions, respectively, and S is the slope of the calibration graphs in mV $decade^{-1}$.

In the matched potential method, the selectivity coefficient was determined by measuring the change in potential upon increasing the primary ion activity from an initial value of a_A to $á_A$ and a_B represent the activity of interfering ion added to the reference solution of primary ion of activity a_A which also brings the same potential change. It is given by the expression:

$$K^{pot}_{A,B} = (á_A - a_A) / a_B$$

In the present studies, a_A and $á_A$ were kept at 1.0×10^{-4} and 1.2×10^{-4} mol L^{-1} hydroxychloroquine sulfate and a_B was experimentally determined.

2. 8. Potentiometric Determination of Hydroxychloroquine Sulfate

The standard addition method was applied [42], in which small increments of the standard solution 1.0×10^{-2} mol L^{-1} of hydroxychloroquine sulfate were added to 50 mL aliquot samples of various concentrations from pure drug or pharmaceutical preparations. The change in milli volt reading was recorded for each increment and used to calculate the concentration of hydroxychloroquine sulfate sample solution using the following equation:

$$C_x = C_s \left(\frac{V_s}{V_x+V_s}\right) \left(10^{\,n\,(\Delta E/S)} - \frac{V_X}{V_x+V_s}\right)^{-1}$$

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

2. 9. Potentiometric Titration of Hydroxychloroquine Sulfate

An aliquots of 1.0×10^{-2} mol L^{-1} drug solution (pure or tablet), were transferred into 50 mL volumetric flasks and made up to the mark with bidistilled water. Different concentrations of hydroxychloroquine sulfate were prepared, then titrated potentiometrically with a standard solution of 1.0×10^{-2} mol L^{-1} NaTPB. The volume of the titrant at equivalence point was obtained using the conventional S-shaped curves. The differential graphs of the titration curves have also been constructed to obtain well defined and sharp end points using the computer program Origin lab.

2. 10. Determination of Hydroxychloroquine Sulfate in Pharmaceutical Preparations

An accurate weight of hydroquine tablets ground and finely powdered in a small Petri dish and dissolved in the bidistilled water up to 30 mL by stirring for 1 h. The solution was filtered in a 50 mL measuring flask. The residue was washed three times with bidistilled water; the volume was completed to the mark by the same solvent to form 1.0×10^{-2} mol L^{-1} solution. The resulting potentials of drug solution were directly measured using CGE electrode.

2. 11. Determination of Hydroxychloroquine Sulfate in Human Urine

Different amounts of hydroxychloroquine sulfate and 5 mL of urine of a healthy person were transferred to a 50 mL measuring flask and completed to the mark using bidistilled water. The contents of the measuring flask were transferred to a 100-mL beaker, and then subjected to standard addition method.

3. Results and Discussion

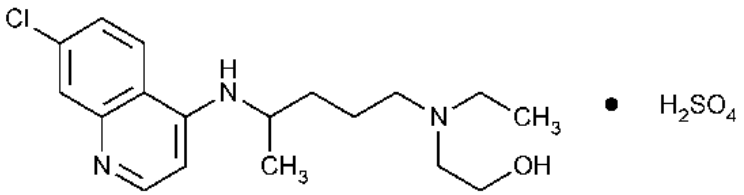

Fig. 1. The chemical structure of hydroxychloroquine sulfate

Hydroxychloroquine cation was found to form 1:2 water insoluble ion-pair with sodium tetraphenylborate as indicated by elemental analysis data and ascertained using conductmetric titration (Fig. 2). The prepared ion-pair was identified and examined in CGE electrode responsive for HCQ cation.

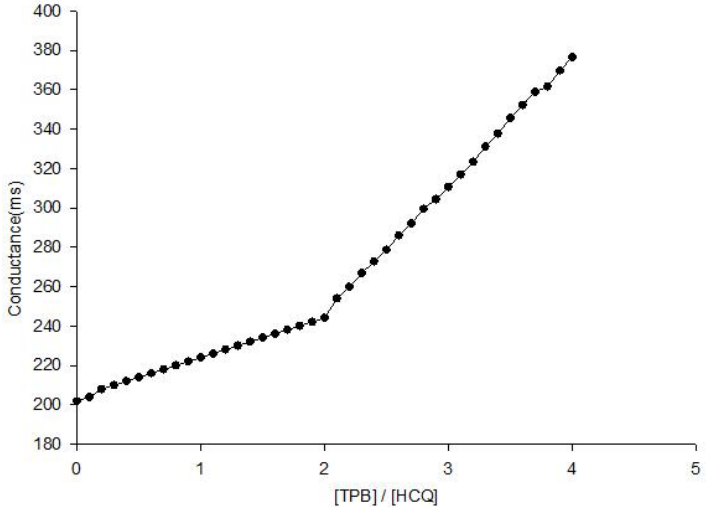

Fig. 2. Conductometric titration curve of 1.0 x 10^{-2} mol L^{-1} HCQS against 1.0 x 10^{-2} mol L^{-1} NaTPB

3. 1. Effect of the Electrode Bed

In the plastic membrane of an ion-selective electrode, the amount of ion-pair should be sufficient to obtain reasonable ionic exchange at the gel layer/test solution interface, which is responsible for the membrane potential. Also, the amount and type of plasticizer should be such that a membrane with good physical properties is produced, which at the same time efficiently acts as a solvent mediator for the ion-pair. A composition of 5.0% (HCQ-TPB) and 47.5% of (DPB) dibutyl phthalate and PVC was used for the preparation of HCQ-TPB coated wire electrode with different conductive beds, namely graphite, copper, silver and aluminium (Table 1).

Table 1. Optimization of the electrode bed and the potentiometric response for coated wire hydroxychloroquine-selective electrodes

Composition of membrane % (w/w; mg)				Slope mV/decade	LR mol L^{-1}	LOD mol L^{-1}	RSD (%)	r^2
Electrode bed	Ion-pair	PVC	DBP					
Graphite	**5**	**47.5**	**47.5**	**30.0±0.2**	**7.1×10^{-5} –1.0×10^{-2}**	**1.8×10^{-5}**	**0.41**	**0.9999**
Silver	5	47.5	47.5	28.5±0.5	1.0×10^{-4}–1.0×10^{-2}	1.0×10^{-4}	2.00	0.9970
Copper	5	47.5	47.5	32.5±0.5	1.0×10^{-4}–1.0×10^{-2}	2.5×10^{-5}	0.81	0.9993
Aluminium	5	47.5	47.5	39.1±0.1	1.0×10^{-4}–1.0×10^{-2}	6.3×10^{-5}	2.10	0.9999

LOD: limit of detection
LR: Linear range
RSD: relative standard deviation (three determinations)
r^2: Correlation Coefficient

After conditioning for 30 minutes, each electrode was examined in the concentration range 1.0×10^{-6} to 1.0×10^{-2} mol L^{-1} of hydroxychloroquine sulfate solution [43]. As can be noticed from Table 1, coated graphite electrode (CGE) showed a good Nernstian slope [30±0.2 mV $decade^{-1}$ at 25±0.1°C] with a concentration range of hydroxychloroquine sulfate (7.1×10^{-5} to 1.0×10^{-2} mol L^{-1} and a detection limit of 1.8×10^{-5} mol L^{-1}) over all the other wires as shown in Fig. 3. This is attributed to the higher electrical conductivity of graphite rod.

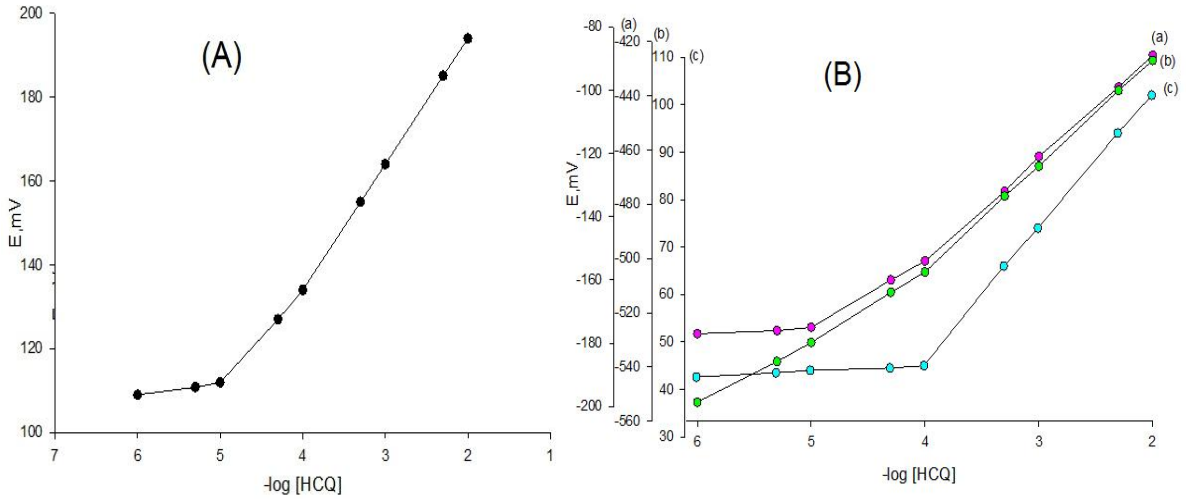

Fig. 3. Calibration curves of (A) coated graphite electrode and (B) (a) coated copper electrode, (b) coated aluminium electrode and (c) coated silver electrode

3.2. Life time of the Electrode

The electrode life time was estimated with the calibration curves, periodical test of a standard solutions and calculating the response slope. The life span of the electrode found to be 20 days during which the electrode, showed a slight gradual decrease in the slope and an increase in the detection limit (Fig. 4). This behavior can be attributed to the decomposition of the ion pair and the loss of other components in the membrane phase that was in contact with the aqueous test solution containing hydroxychloroquine cation. It was observed that the life time of the CGE found to be short. This would be due to the formation of water film at the interface between the membrane and the graphite and interaction of the membrane species with the graphite.

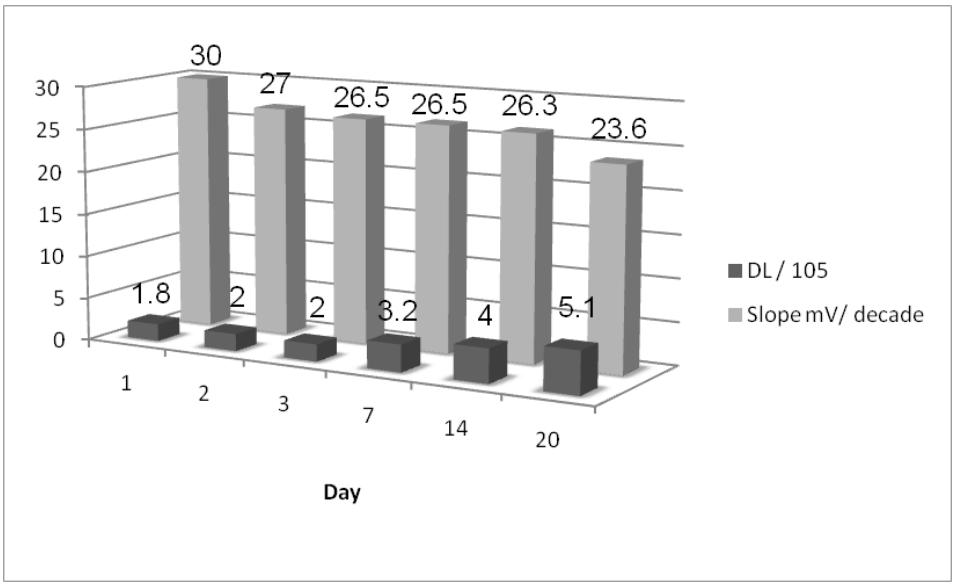

Fig. 4. Behavior of CGE electrode in term of detection limit and slope during 20 days

3. 3. Regeneration of the Electrode

The regeneration of the electrode was tried simply by reformation of the ion-pair on the external gel layer of membrane as reported previously [44]. The regeneration of the hydroxychloroquine sulfate membrane was successfully achieved by soaking the expired electrode for 24 h in a solution that was $1.0x10^{-2}$ mol L^{-1} sodium tetraphenylborate, followed by soaking for 3 h in $1.0x10^{-2}$ mol L^{-1} hydroxychloroquine sulfate. The calibration graph for exhausted showed (slope 23.6±0.6 mV $decade^{-1}$) and after regeneration (slope 27.0±0.3mV $decade^{-1}$). It was found that the life span of the regenerated electrode is limited to 8 h due to the ease of leaching of the lipophilic salts from the gel layer at the surface of the electrode compared with those that are attached homogeneously to the membrane network through the solvent mediator.

3.4. Dynamic Response Time and Repeatability of the Electrode

The dynamic response time [45], is defined as the time which elapses between the instant at which an ion-selective electrode and a reference electrode (ISE cell) are brought into contact with a sample solution. The dynamic response time of the electrode was tested by measuring the time required to achieve a steady-state potential (within ±1 mV) after successive immersions of the electrode in a series of drug solutions, each having a 10-fold increase in concentration from 1.0×10^{-5} to 1.0×10^{-2} mol L^{-1}. The electrode yielded a steady potential within 10 s as shown in Fig. 5.

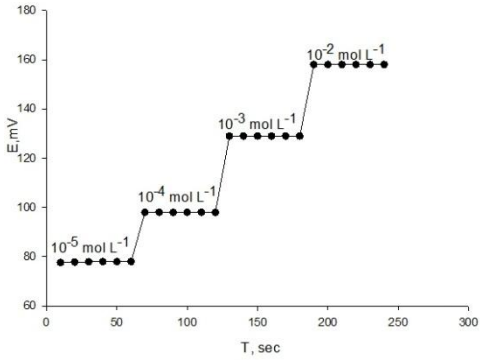

Fig. 5. Dynamic response time of hydroxychloroquine CGE electrode for step change in concentrations of hydroxychloroquine sulfate from low to high

The repeatability of the potentials readings for the electrode was examined by subsequent measurement in 1.0×10^{-3} mol L^{-1} hydroxychloroquine sulfate solution immediately after measuring in 1.0×10^{-2} mol L^{-1} hydroxychloroquine sulfate solution (Fig. 6). An insignificant difference in potential readings was obtained, indicating good repeatability of the constructed electrode.

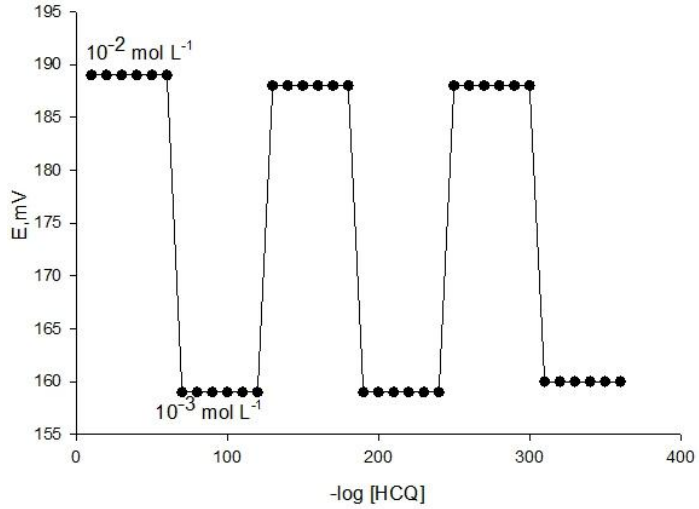

Fig. 6. Dynamic response of hydroxychloroquine CGE electrode for several high-to-low sample cycles

3.5. Effect of the pH

The effect of the pH of the solution on the response of the proposed electrode was studied for two concentrations of hydroxychloroquine sulfate (1.0×10^{-3} and 1.0×10^{-4} mol L^{-1}) in the pH range of 1.5–11.0. The pH was adjusted with (0.1-1.0 mol L^{-1}) solutions of hydrochloric acid or sodium hydroxide. The results obtained show that, the potential response remains almost constant over the pH range 2-7 which can be taken as the working pH range of the electrode (Fig. 7). Above this pH range, the potential showed a sharp decrease due to the formation of the nonprotonated HCQ, leading to a decrease in concentration of HCQ^{+}. The decrease in the potential at pH lower than 2 may be due to interference of hydronium ion and penetration of H_3O^{+} into the membrane surface or a gradual increase of protonated species and dependence of the e.m.f. on the pH of the solution.

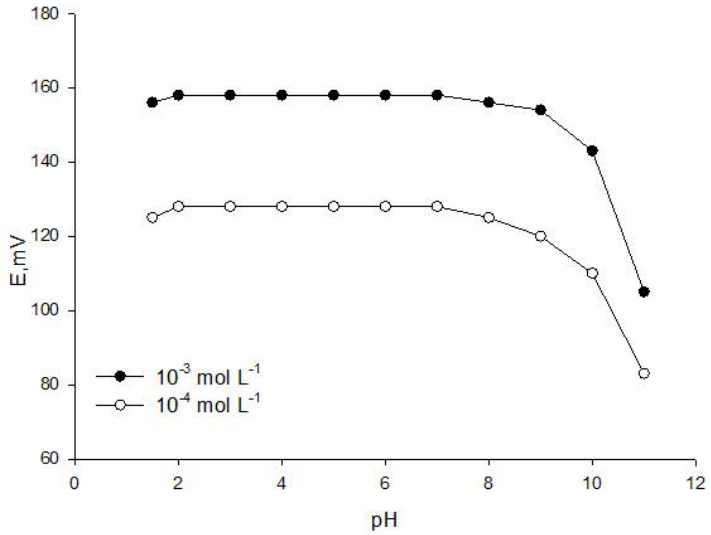

Fig. 7. Effect of pH of the test solution on the potential response of hydroxychloroquine CGE electrode

3.6. Selectivity of the Electrode

The selectivity coefficients for hydroxychloroquine sulfate electrode with respect to a variety of inorganic cations, vitamins, sugars and amino acids were determined by the separate solution method (SSM) and the matched potential method (MPM). The selectivity coefficients values of the electrode (Table 2) reflect a very high selectivity of the investigated electrode for the HCQ cation. The inorganic cations do not interfere owing to the differences in ionic size and consequently their mobilities and permeabilities as compared with HCQ^+. The selectivity sequence significantly differs from the so called Hofmeister selectivity sequence [46] (i.e. selectivity solely based on lipophilicity of cation). In case of non-ionic species, the high selectivity is mainly attributed to the difference in polarity and to the lipophilic nature of their molecules relative to hydroxychloroquine cation. The mechanism of selectivity is mainly based on the stereospecificity and electrostatic environment, and is dependent on how much fitting is present between the locations of the lipophilicity sites in two competing species in the bathing solution side and those present in the receptor of the ion-exchanger [47].

Table 2. Selectivity coefficient values of the hydroxychloroquine sulfate-selective electrode at 25°C

	$k_{Prx,j}^{pot}$	
Interferent	SSM	MPM
K^+	6.3×10^{-3}	-
NH_4^+	7.9×10^{-3}	-
Li^+	5.0×10^{-3}	-
Fe^{2+}	1.6×10^{-4}	-
Ca^{2+}	5.0×10^{-4}	-
Mg^{2+}	3.2×10^{-4}	-
Mn^{2+}	4.0×10^{-4}	-
Cu^{2+}	2.5×10^{-3}	-
Co^{2+}	2.5×10^{-4}	-
Vitamine C	-	2.5×10^{-3}
Vitamine B_6	-	3.2×10^{-3}
Glucose	-	1.0×10^{-2}
Fructose	-	1.0×10^{-2}
Lactose	-	2.5×10^{-3}
Maltose	-	2.5×10^{-3}
Urea	-	7.9×10^{-3}
Glycine	-	7.9×10^{-3}
β-alanine	-	1.0×10^{-2}
Hestidine	-	6.3×10^{-2}

3.7. Effect of Temperature

Calibration plots were constructed in the test solution temperatures (15, 20, 25, 30, 40, and 50 °C) for CGE electrode. For the determination of the thermal coefficients (dE°/dt) of the cells, the standard electrode potentials E°, obtained from the calibration graphs as the intercepts at pHCQS = 0, were plotted versus (t − 25), where t is the temperature (°C) of the test solution. A straight line is obtained according to the Antropov's equation [48]

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

The slope of the straight line obtained represents the thermal coefficient of the electrode, amounting to 0.0011 mV $^{\circ}C^{-1}$. This low value of thermal coefficient reveals a good thermal stability of the electrode within the studied temperature range (15–50 $^{\circ}C$).

3.8. Analytical applications

The investigated electrode was successfully used for the potentiometric determination of hydroxychloroquine sulfate in pure solutions, pharmaceutical preparations (Hydroquine® tablets) and spiked urine samples by applying standard addition method. The obtained average recovery and relative standard deviation values are summarized in Tables 3and 4, which reflect the high accuracy and precision of the electrode.

Table 3. Determination of hydroxychloroquine sulfate in pure solutions and pharmaceutical preparations applying the standard addition and the potentiometric titration methods

	Standard addition method			Potentiometric titration method		
Sample	Taken (mg)	Recovery (%)	RSD (%)	Taken (mg)	Recovery (%)	RSD (%)
Pure sample	0.43	99.9	2.10	8.68	100.0	0.90
	1.08	100.6	0.73	17.36	101.3	1.30
	2.17	100.2	1.67	26.04	100.0	0.89
	4.34	99.0	1.80			
Hydroquine	0.43	98.3	0.88	8.68	100.0	0.82
(200mg/tablet)	1.08	99.2	2.10	17.36	98.8	0.88
sample	2.17	99.4	1.37	26.04	100.8	0.23
	4.34	100.0	1.90			

Table 4. Determination of hydroxychloroquine sulfate in spiked urine samples applying the standard addition method

Sample	Taken (mg)	Recovery (%)	RSD (%)
Urine	0.43	101.3	1.10
	1.08	99.1	2.00
	2.17	100. 1	0.87
	4.34	100.0	1.80

In addition, CGE electrode is used as an indicator electrode in potentiometric titration of HCQS with NaTPB in pure solutions and pharmaceutical preparations (Table 3). It is clear that the amount of HCQ ion can be accurately determined with this electrode (Fig. 8).

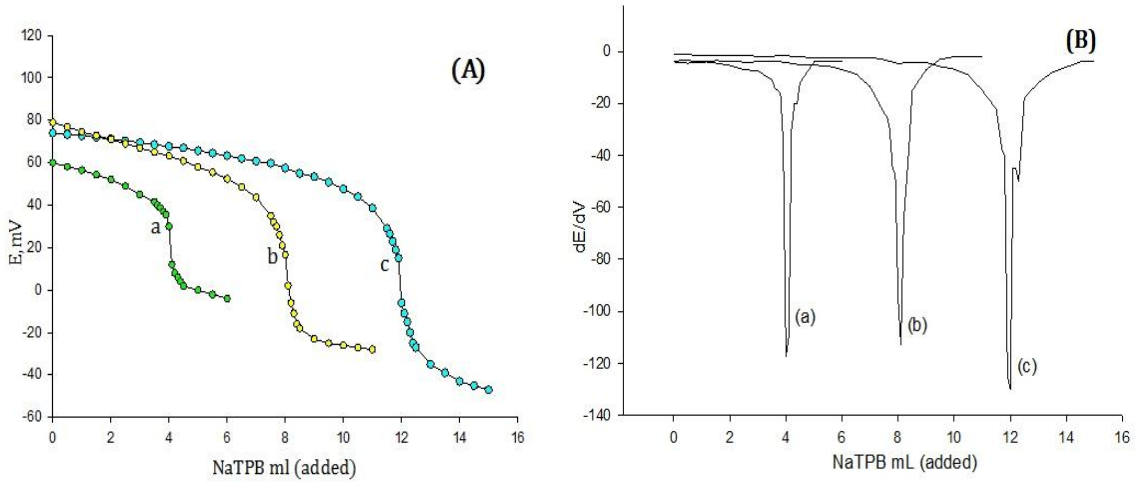

Fig. 8. (A) Potentiometric titration curves of (a) 2 (b) 4 and (c) 6 mL of 10^{-2} mol L^{-1} hydroxychloroquine sulfate using CGE electrode and 10^{-2} mol L^{-1} NaTPB as titrant and (B) its first order derivative

3.9. Statistical analysis and validity of the proposed method

The linearity, limit of detection, precision, accuracy, and ruggedness/robustness were the parameters used for the method validation. As mentioned before, the measuring range of the hydroxychloroquine electrode is between 7.1×10^{-5} and 1.0×10^{-2} mol L^{-1} hydroxychloroquine sulfate

3.9.1. Ruggedness

For ruggedness of the method a comparison was performed between the intra- and inter-day assay results for hydroxychloroquine sulfate obtained by two M. Sc. students. The RSD values for the intra- and inter-day assays of hydroxychloroquine sulfate in the cited formulations performed in the same laboratory by the two analysts did not exceed 2.10% which indicates that the method is capable of producing results with high precision.

3.9.2. Robustness

The robustness was examined while the parameter values (pH of the medium and the laboratory temperature) were being deliberately slightly changed. Hydroxychloroquine sulfate recovery percentages were good under most conditions, not showing any significant change when the critical parameters were modified.

The results obtained from the standard addition method of the drug were compared with those obtained from the potentiometric titration method by applying F-and t-tests [49]. The results (Table 5) show that the calculated F- and t-values did not exceed the theoretical values, reflecting the accuracy and precision of the applied method.

Table 5. Statistical comparison between the results of an analysis of a pharmaceutical preparation applying the standard addition and potentiometric titration methods

Parameters	Standard addition method	Potentiometric titration method
Mean recovery (%)	99.3[a]	99.9[b]
SD	0.71	1.01
RSD (%)	0.71	1.01
F-ratio	2.04 (9.55) [c]	
t-test	1.04 (2.57) [d]	

a: Average of four determinations
b: Average of three determinations
SD: standard deviation
RSD: relative standard deviation
c: Tabulated F-value at 95% confidence level
d:Tabulated t-value at 95% confidence level and five degrees of freedom

3.10. Comparison with reported methods

The performance characteristics of the proposed electrode and those of the reported methods are compiled in Table 5 for comparison. It is clear that the detection limit of the constructed CGE electrode is lower than that of pulse differential voltammetry method [20]. In addition, the range of concentration of CGE electrode is wider than that of both pulse differential voltammetry and spectrophotometry methods [20], which indicates the ability of the constructed electrode to face such automated methods.

Table 6. Comparison of the proposed hydroxychloroquine ion-selective electrode method with published methods

Method	Linear range mol L^{-1}	LOD mol L^{-1}	r^2	RSD (%)	Ref
Spectrophotometry	4.0×10^{-6}-2.0×10^{-5}	2.30×10^{-8}	0.9999	0.36	[20]
Pulse differential voltammetry	2.0×10^{-5}-5.0×10^{-4}	2.60×10^{-5}	0.9999	0.46	[20]
Ion Selective Electrode					
CGE	7.1×10^{-5}–1.0×10^{-2}	1.8×10^{-5}	0.9999	0.41	[P.S]

LOD: limit of detection
r^2: correlation coefficient
RSD: relative standard deviation (three determinations)
Ref.: reference
P.S.: present study

4. Conclusion

The proposed electrode based on HCQ-TPB as the electroactive compound can be used as an interesting alternative analytical tool for the determination of hydroxychloroquine in pure solutions, pharmaceutical preparations and human urine samples. The electrode showed a Nernstian slope of 30.0±0.2 mV $decade^{-1}$, a wide concentration range from 7.1×10^{-5} to 1.0×10^{-2} mol L^{-1} , a low detection limit of 1.8×10^{-5} mol L^{-1} and a short response time (≤ 10 s) over the pH range 2.0-7.0. The proposed potentiometric method offers the advantages of simplicity, accuracy and applicability to turbid and coloured sample solutions.

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