Circumventing traditional conditioning protocols in polymer membrane-based ion-selective electrodes

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ABSTRACT: Preparation of ISEs often requires long and complicated conditioning protocols limiting their application as tools for in field measurements. Herein, we eliminated the need for conditioning by loading the membrane cocktail with primary ion solution. This protocol significantly shortens the preparation time of ISEs yielding functional electrodes with submicromolar detection limits.

The scientific research in ion-selective electrodes (ISEs) has gained momentum within the last years due to improvements in the limits of detection and selectivity, becoming now applicable for trace-level measurements by understanding transmembrane ion fluxes.[1] The response of ISEs can be described by the phase boundary potential, E_{PB}, according the following equation:

$$E_{PB} = E^0 + \frac{RT}{z_I F} ln \frac{a_{I\,(aq)}}{a_{I\,(org)}} \qquad (1)$$

Here $a_{I\,(aq)}$ and $a_{I\,(org)}$ are the activities of primary ion (I) of charge z in aqueous and organic phases respectively, while E^o, R, T, and F are the standard potential, gas constant, temperature and Faraday constant, respectively. When $a_{I\,(org)}$ is kept constant, the equation 1 reduces to the well-known Nernst equation:

$$E_{PB} = E^0 + \frac{RT}{z_I F} ln\, a_{I\,(aq)} \qquad (2)$$

In order to render an ion-selective membrane functional, the ionophore and lipophilic ionic sites are required. One of the major roles of ionophore is to make relatively strong complexes with the primary ion, thereby establishing their constant activity in the membrane.[2] For more details see Equations SI1-SI5 in the supporting information. The role of the lipophilic ionic sites is to provide ion-exchange properties. For cation selective membrane, this process could be described by the following equilibrium:

$$a^{zI+}_{I(aq)} + nL_{org} + M^+_{org}R^-_{org} \rightarrow [IL_n]^{zI+}_{org} + M^+_{aq} + R^-_{org} \qquad (3)$$

where L is a ligand (ionophore) that forms ion-ionophore complex with ion I of stoichiometry n. $M^+_{org}R^-_{org}$ is a lipophilic ion exchanger composed of lipophilic anion R^- and its counterion M^+. Partitioning of I from aqueous sample into the membrane results in its exchange with M^+. Anion R^-_{org} remains in the membrane thereby rendering the membrane permselective while preserving the charge balance.[3] In a typical experimental protocol for the preparation of ion-selective membranes the ion-exchange process is obtained by conditioning (soaking) the membrane in an aqueous solution containing the ion I (traditional protocol).[4]

Significant effort in ISEs field has been spent on researching ways to miniaturize[5–9] and optimize/simplify the preparation of ISEs.[10–12] Reducing or eliminating the need for the conditioning step prior to the use of the electrodes is an important step for devising a simple, practical protocol for ISEs applications.[12] This would enable non-trained personnel to use ISEs quickly and reliably. In this work we propose a simple alteration of the sensor's conditioning protocol. Instead of placing the ISEs in a solution of primary ions I, solution is added directly into the membrane cocktail prior to its casting. The concentration of that solution is calculated to allow for stoichiometric exchange of I and M. Consequently, ions I are present in the membrane facilitating the formation of ion-ionophore complex according to the following equilibrium:

$$I^{zI+}_{org} + nL_{org} + R^-_{org} \rightarrow [IL_n]^{zI+}_{org} + R^-_{org} \qquad (4)$$

Therefore, upon casting, the membrane is already preloaded with complexed ions I in the form of the ion-ionophore complex $[IL_n]_{org}^{zI+}$. Satisfaction of stoichiometry implies that the concentration of the complex in the membrane is constant thus fulfilling the requirements for application of Equation 2. The well-formed Nernstian responses are therefore expected for these non-conditioned ISEs.

EXPERIMENTAL

Reagents

4-*tert*-Butylcalix[4]arene-tetraacetic acid tetraethyl ester (sodium ionophore X), *o*-xylylenebis(*N*,*N*-diisobutyldithiocarbama-te) (copper(II) ionophore I), potassium tetrakis[3,5 bis(trifluorome-thyl) phenyl]borate (KTFPB), sodium tetrakis[3,5 bis(trifluoro-methyl) phenyl]borate (NaTFPB), tridodecylmethylammonium nitrate ($TDMANO_3$), bis(2-ethylhexyl) sebacate (DOS), high molecular weight poly(vinyl) chloride (PVC), and tetrahydrofuran (THF), all of Selectophore grade, were purchased from Sigma-Aldrich. [9]Mercuracarborand-3 (MC3) was synthesized in house as described previously.[14] The salts $AgPF_6$, $AgNO_3$, $NaNO_3$, NaI, and KCl were also obtained from Sigma-Aldrich. All aqueous solutions were prepared in ultra-pure water obtained with Purelab Ultra water purification system (resistance 18 MΩ cm).

Membranes

The Na^+-selective membrane contained 10.0 mmol kg^{-1} of sodium ionophore X and 5.0 mmol kg^{-1} of NaTFPB. The Ag^+-selective membrane contained 10.0 mmol kg^{-1} of copper (II) ionophore I and 5.0 mmol kg^{-1} of NaTFPB.[15] The I^--ISEs contained 1.0 mmol kg^{-1} of MC3 and 0.75 mmol kg^{-1} of $TDMANO_3$. Ion-selective membranes were prepared by dissolving the above-mentioned components together with PVC (33.3 wt %) and DOS (66.6 wt %) in 2 mL THF and the resulting cocktail was vortexed for 30 min.

To avoid conditioning steps, 25 µL of 0.1 M $AgNO_3$ solution was added to the Ag^+-selective membrane and 1.8 µL of 0.1 M NaI solution was added to the I^--selective membrane before vortexing. No additional solution was added for the Na^+-selective membrane. Control membranes were also prepared: For Ag^+-selective ISEs, no $AgNO_3$ solution was added, for I^--selective control membranes, no NaI solution was added. For Na^+-selective electrodes, KTFPB was used as ion-exchanger instead of NaTFPB. The resulting membrane cocktail was cast into a glass ring (26 mm inner diameter) secured to a glass plate, and the solvent was allowed to evaporate overnight. This resulted in a membrane film with a thickness of approximately 200 ± 10 µm (obtained using caliper).

Electrodes

The ISEs were prepared as follows: Firstly, a 3.2 mm diameter disk was cut from the parent membrane film and adhered to the end of a PVC tube (2 cm length, 1.6 mm i.d.) using THF. The other end of the tube was fixed to a 1 mL pipette tip that held the inner filling solution. Inner filling solutions for the Na^+-ISE, Ag^+-ISE, and I^--ISE were 1.0×10^{-3} M $NaNO_3$, 1.0×10^{-3} M $AgNO_3$ and 1.0×10^{-3} M NaI, respectively. A diaphragm separated the internal filling solutions from the reference half-cell (Ag/AgCl in 0.1 M KCl).

EMF measurements

Potential measurements of the non-conditioned ISEs began immediately after adding the inner filling solution to the electrode. Measurements were carried out in 100 mL polypropylene beakers. A high-input impedance (10^{15} Ω) EMF-16 multichannel data acquisition system (Precision Electrochemistry EMF Interface, Lawson Laboratories) was used to monitor the potentials, at room temperature (22 ºC) and under constant stirring. A double junction Ag/AgCl/3M KCl/1M LiOAc (Metrohm AG) was used as the reference electrode. All EMF values were corrected for liquid-junction potentials according to the Henderson equation, and activity coefficients were calculated by the Debye-Hückel approximation.

Selectivity measurements

ISEs for selectivity measurements were prepared according to the protocol described in 'Membranes and Electrodes' with some modifications. For Na^+-ISEs, lipophilic ion exchanger - KTFPB was used instead of NaTFPB. Ag^+ cocktail solution was loaded with 5 mmol kg^{-1} of $NaNO_3$ while 5 mmol kg^{-1} of KNO_3 were added into I^- membrane solution. Inner filling solutions for the Na^+-ISE, Ag^+-ISE, and I^--ISE were 1.0×10^{-3} M KNO_3, 1.0×10^{-3} M $NaNO_3$ and 1.0×10^{-3} M KNO_3, respectively. Responses towards all ions were recorded according to separate solution method as described by Bakker.[16]

RESULTS AND DISCUSSION

Response characteristics

The establishment of the ion-ionophore complex equilibrium is key to create the potential gradient at the interface of the electrode while maintaining constant activity of analyte ions inside the membrane.[17] By understanding, the ion flux at the membrane/sample interface, it enables the ion-selective membrane composition to be optimized while avoiding long conditioning steps. The equilibrium between complexed and free ionophore in the membrane could be obtained during the preparation of the membrane by adding the ion of interest directly to the membrane cocktail at stoichiometrically optimized amount. In the preparation step, consideration should be given to the lipophilicity of the counterion of the loading solution (see the Supporting Information Figure S1 and accompanying discussion).

Thus, Figure 1A shows the calibration curve of the non-conditioned Na^+-ISE that contains NaTFPB as the ion-exchanger. This calibration curve displays a Nernstian slope of 57.05 ± 2.03 mV decade^{-1} and a submicromolar limit of detection of $3.24 \times 10^{-7} \pm 0.02$ M, showing a similar performance as the conditioned Na^+-ISEs previously reported using a similar membrane composition.[18] Conversely, the calibration curve of the non-conditioned Na^+-ISE control membranes containing KTFPB instead of NaTFPB as the ion-exchanger (Figure 1B) demonstrated a super-Nernstian behavior upon initial exposure to sodium ions. This is caused by a flux of sodium ions from the sample solution into the bulk of ion-selective membrane as previously reported in the literature.

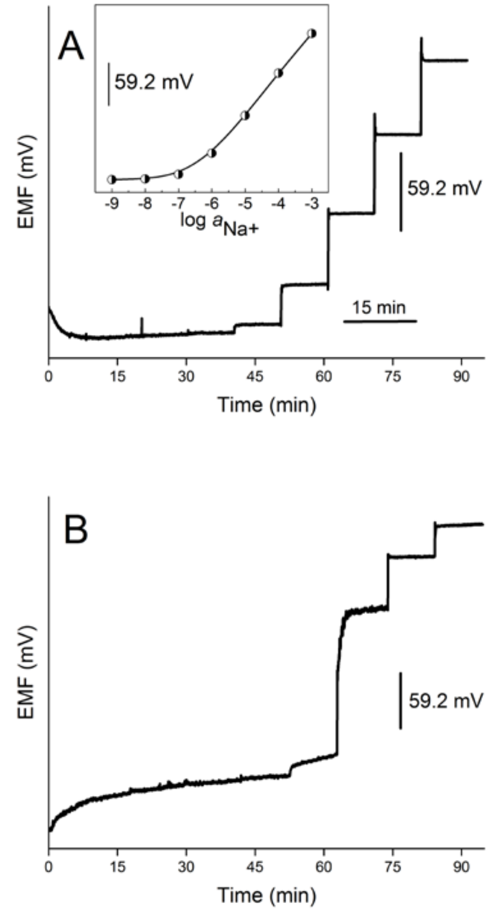


Figure 1. Trace lines for the non-conditioned Na^+-ISE containing A) NaTFPB and B) KTFPB as the ion-exchanger. Inset in Figure 1A displays the respective calibration curve. All other components of the membranes in A and B are the same.

Similarly, the calibration curve (Figure 2A) of the non-conditioned Ag^+-ISE with preloaded $AgNO_3$ solution also displays a Nernstian slope of 58.05 ± 0.77 mV decade^{-1} and a limit of detection of $2.69 \times 10^{-7} \pm 0.03$ M, that are similar to the traditional conditioned Ag^+-ISE as reported in the literature, but with higher limit of detection.[19] The small discrepancy in the observed and reported detection limits could be caused by the trans-membrane ion flux from the inner filling solution in the direction of the sample.[20] This could disturb the local equilibrium at the sample/membrane interface consequently worsening the detection limit of ISEs.

On the other hand, a super-Nernstian response was observed (Figure 2B) for the non-conditioned Ag^+-ISE in which no $AgNO_3$ solution was added to the cocktail solution (control membrane). Similarly to the Na^+-ISE the super-Nernstian response was also observed due to the initial exposure of ISEs to the primary ions (silver ions). In addition, control membranes were subsequently conditioned for 2, 4, 6, 8, 10, 12 and 24 h in 1.0×10^{-3} M of $AgNO_3$ solution and a minimum of 12 h was required to obtain ISEs with Nernstian response slopes (data not shown). Moreover, traditionally prepared Ag^+-ISEs exhibited similar response characteristics (lower detection limits and Nernstian behavior, Figure S2) to those conditioned according to the proposed methodology. This demonstrates the practicality of adding the solution of primary ions into the membrane cocktail to further reduce the preparation time of ISEs.

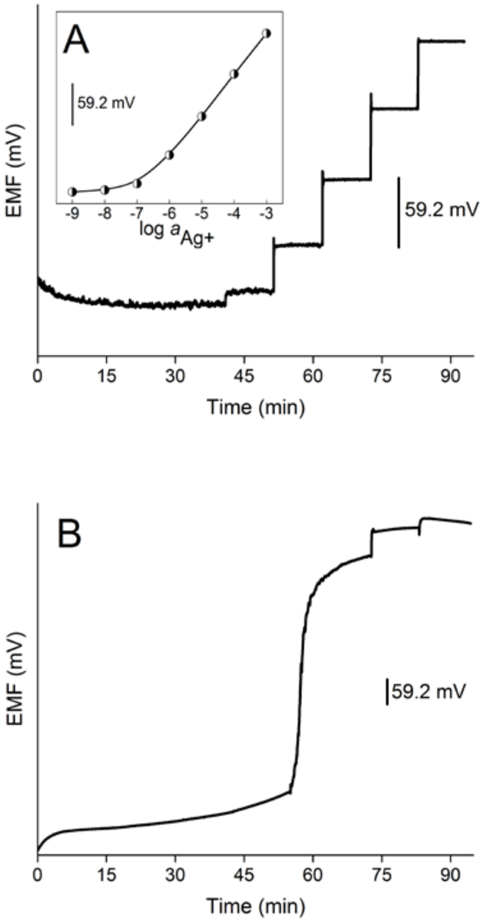
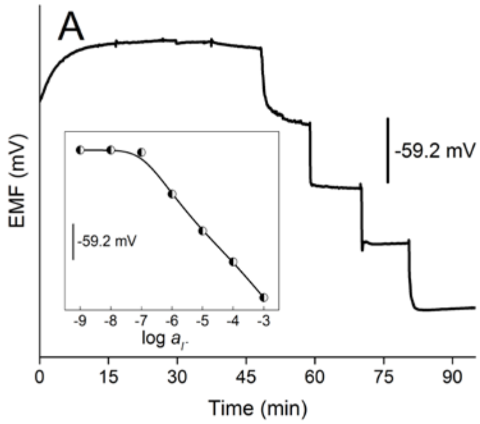


Figure 2. Trace lines for the non-conditioned Ag^+-ISEs containing A) $AgNO_3$ solution and B) without $AgNO_3$ solution in

the membrane. Inset in 2A displays the respective calibration curve. All other components of the membranes in A and B are the same.

The Nernstian behavior of the non-conditioned cation-selective system that was doped with the primary ion during the membrane preparation for sodium and silver ions were also tested for an anion-selective system. As described in the experimental part the I^--ISEs were prepared with and without NaI in the membrane composition. As shown in Figure 3A, the calibration curve of the non-conditioned I^--ISEs, where NaI solution was added into the cocktail, presents a Nernstian slope of 57.59 ± 2.56 mV decade^{-1} and a limit of detection of 8.51 x 10^{-8} ± 0.07 M. This is in good agreement with the traditional conditioned I^--ISEs based on the same ionophore as reported in the literature, but the observed detection limit was higher by one order of magnitude.[21] The non-optimal behavior of MC-3 based ISEs at the lower detection end could be caused by the strong interference from the hydroxide ions.[22] Similar as in cation system, when a solution of the analyte was not added into the cocktail solution (blank membranes for iodide) a super-Nernstian response was observed (Figure 3B).

The non-conditioned ISEs were also characterized in terms of selectivity using an unbiased method. Therefore, each electrode was conditioned in a solution of less discriminated interfering ion prior to the performed measurements.[16] Table 1 shows the selectivity values obtained for Na^+-ISE, Ag^+-ISE and I^--ISE.



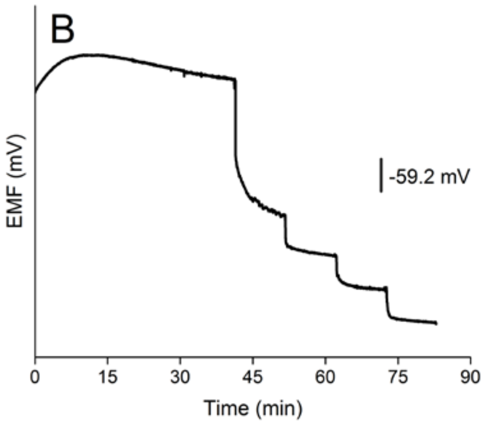


Figure 3. Figure 3. Trace lines for the non-conditioned I^--ISEs containing A) NaI solution and B) and without NaI solution in the membrane. Inset in 3A displays the respective calibration curve. All other components of the membranes in A and B are the same.

Table 1. Unbiased selectivity coefficients obtained for the Na^+-ISEs (slope: 50.23 ± 0.09 mV decade^{-1}), Ag^+-ISEs (slope: 57.32 ± 3.27 mV decade^{-1}) and I^--ISEs (slope: - 56.33 ± 0.17 mV decade^{-1}) with corresponding slope values for interfering ions.

Ion J^{Z+}	$K^{pot}_{Na,J}$	Slope (mV decade^{-1})
K^+	-1.1 ± 0.1	56.35 ± 0.13
H^+	-4.5 ± 0.3	51.61 ± 1.80
Li^+	-5.2 ± 0.3	53.03 ± 1.39
Ion J^{Z+}	$K^{pot}_{Ag,J}$	Slope (mV decade^{-1})
Na^+	-6.9 ± 1.2	53.11 ± 1.15
K^+	-6.4 ± 0.9	54.51 ± 0.68
H^+	-7.6 ± 0.5	50.58 ± 3.23
Cu^+	-8.7 ± 0.6	59.91 ± 0.61
Ion J^{Z+}	$K^{pot}_{I,J}$	Slope (mV decade^{-1})
Br^-	-1.3 ± 0.0	-64.71 ± 2.92
Cl^-	-1.3 ± 0.0	-64.65 ± 2.71
NO_3^-	-2.1 ± 0.0	-55.93 ± 4.00

CONCLUSIONS

In this work, new methodology for the preparation of ISEs with reduced equilibration time is proposed. This was achieved by adding a small aliquot of primary ions into the cocktail solution giving rise to ISEs with submicromolar detection limits. This facilitates the use of ISEs in the field for non-trained personnel. Further characterization has to be carried out to fully understand the properties on the sensor prepared according to the proposed modified protocol.

ASSOCIATED CONTENT

Supporting Information

Further experimental details. This material is available free of charge via the Internet at http://pubs.acs.org."

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All The manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript.

Notes

The authors declare no competing financial interests.

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