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Interpretation of chronopotentiometric transients of ion-selective membranes with two transition times

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Abstract

Passing currents through ion-selective membranes has contributed to the development of a variety of novel methods. In this work, chronopotentiometric (CP) transients with two transition times (breakpoints) are presented for the first time, with the theoretical interpretation of such voltage transients. The validity of our theory has been confirmed in experiments utilizing ETH 5294 chromoionophore-based pH sensitive membranes with and without lipophilic background electrolyte and ETH 5234 ionophore-based calcium selective membranes in which the ionophore forms 3:1 complexes with Ca^{2+} ions. The conditions under which two breakpoints can be identified in the chronopotentiometric voltage transients are discussed.

Spectroelectrochemical microscopy (SpECM) is used to show that the two breakpoints in the CP curves emerge approximately when the free ionophore and ion-ionophore complex concentrations approach zero at the opposite membrane-solution interfaces. The two breakpoint times can be utilized to follow simultaneously the concentration changes of the free ionophore, the ion-ionophore complex, and the mobile anionic sites in cation-selective membranes. In membranes with known composition, the time instances where breakpoints occur can be used to estimate the free ionophore and the ion-ionophore complex diffusion coefficients.

INTRODUCTION

Cation-selective liquid membranes are extensively used for the determination of ion concentrations in clinical, environmental, and other complex samples. These membranes usually contain (1) an ionophore, e.g., the H^+ -selective chromoionophore I (ETH 5294), (2) mobile or fixed ion-exchanger sites, e.g., sodium tetrakis[3,5-bis(trifluoromethyl) phenyl] borate (NaTFPB), and sometimes (3) a lipophilic background electrolyte to reduce the membrane resistance, e.g., tetradodecylammonium tetrakis(4-chlorophenyl)borate (ETH 500). The ionophore provides the membrane its selectivity towards a specific cation, while the ion-exchanger sites make the membrane permselective. Although ion-selective membranes are mainly used for zero-current equilibrium measurements, a variety of non-equilibrium

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techniques have been devised to probe membrane characteristics or adjust membrane responses. For example, from the transition time (the instance of the breakpoint or breakpoint time) that appears in chronoamperometric (CA) [1–4] and chronopotentiometric (CP) [5–7] transients, the concentration or the diffusion coefficient of the free ionophore inside the membrane has been determined.

In CA and CP experiments with cation-selective membranes, cations are forced into the membrane at its positively polarized side. These ions are complexed by the ionophore in the membrane. As a result of this complexation reaction, the concentration of the free ionophore decreases and the concentration of the ion-ionophore complex increases at the membrane side of the phase boundary. A breakpoint emerges in the CA and CP transients at a transition time when the free ionophore concentration approaches zero at the membrane boundary. It has been shown [1–3,5–7] that the transition time is proportional to the diffusion coefficient and the square of the free ionophore concentration so that it can be used to determine either of these parameters from Eq. 1.

$$\tau_{1/2} = \frac{FAR_{\text{bulk}}C_{\text{L,free}}\sqrt{D_{\text{L,free}}}\pi}{2V} = \frac{FAC_{\text{L,free}}\sqrt{D_{\text{L,free}}}\pi}{2I} \quad (1)$$

where F is the Faraday constant, A is the surface area of the membrane, $C_{\text{L,free}}$ and $D_{\text{L,free}}$ are the concentration and the diffusion coefficient of the free ionophore in the membrane, respectively, R_{bulk} is the bulk resistance of the membrane, V is the applied voltage in CA experiments, and I is the applied current in CP experiments.

For example, the CA method provided a unique possibility to trace the leaching-related loss of the ionophore from ion-selective membranes *in vivo* and estimate the residual lifetime of the membrane [2]. In membranes with constant composition, the CA method has been used to determine the diffusion coefficients of a variety of ionophores [1,3]. Recently, it was shown that chronopotentiometry can be used in the same way, with substantial theoretical and experimental advantages [5–7]. The theoretical advantage is related to the validity of the constant current assumption in the chronoamperometric theory. In contrast to chronoamperometry, this assumption is not necessary in chronopotentiometry since the galvanostatic mode guarantees the constant current. The experimental advantage is that the determination of the transition time (required for the calculation of the diffusion coefficient or the membrane composition) is generally simpler in the CP voltage-time transients than in the CA current-time curves because the breakpoints are more easily discerned, i.e., the change in the slope of the curves at the breakpoint time is larger.

The theoretical description of the CP transient has revealed that for some membranes the ion-ionophore complex boundary concentration can approach zero on the negative side of the membrane before the free ionophore concentration approaches zero concentration on the positive side [5–7]. For these membranes, the breakpoint in the transient is related to the ion-ionophore concentration in the membrane, so that the ion-ionophore complex concentrations or the diffusion coefficient of the ion-ionophore complex or anion can be determined from the transition time.

In this work, we show that under certain experimental conditions two breakpoints emerge in the CP voltage transients. One of the breakpoints can be correlated to the concentration polarization of the free ionophore while the other is correlated to the concentration polarization of the ion-ionophore complex. To verify the correlation between the breakpoint times and the times when the concentrations of the free and the complexed ionophores approach zero concentration (the zero concentration times) at the opposite membrane boundaries,

spectroelectrochemical microscopy (SpECM) has been used in this work [8–10]. SpECM is a technique that allows the simultaneous measurement of the voltage transient and the concentration profiles of the free and complexed ionophores (chromoionophores) inside the membrane [7,9].

From the two breakpoint times, both the free ionophore and the ion-ionophore complex concentrations can be determined in a single experiment. In addition, the two breakpoint times can be used for the simultaneous determination of the diffusion coefficients of the free ionophore and the ion-ionophore complex or lipophilic anion. The determination of diffusion coefficients in ISE membranes [3,5–7] has gained importance with the realization that the detection limit of ISEs is commonly limited by primary ion fluxes through the membrane [11,12]. The magnitude of these fluxes depends in part on the diffusion coefficient of the ion-ionophore complex inside the membrane. By minimizing or eliminating these fluxes, sub-nanomolar detection limits have been achieved [11,13–17]. In addition to the CP determination of the breakpoint times, understanding the mechanism of processes inside the membrane during applied currents is also important in other CP applications, e.g., (i) in pulsed potentiometric methods that allowed the detection of polyions [18]; (ii) in pulsed potentiometric applications that improved the selectivity of calcium sensors [19]; and (iii) during the coulometric generation of ions from the ion-selective membranes [20,21]. In these CP experiments the membranes are commonly loaded with a high concentration of lipophilic background electrolyte to reduce membrane resistance and avoid migration effects. Consequently, the CP transients of such membranes were studied most frequently [20]. In this paper, we focus on the voltage transients of ion-selective membranes cast without a background electrolyte, and propose a new interpretation for multiple transition times that can emerge in CP experiments. The new interpretation is valid for membranes cast with or without a background electrolyte.

THEORY

The concentration profiles of the free ionophore and ion-ionophore complex in the bulk membrane cross-section are approximately flat at zero current [9,22]. However, when a current is applied, concentration gradients form because ions are forced into the membrane on one side and out of the membrane on the other side. The concentration profiles in neutral ionophore-based cation-selective membranes during an applied current step have been described previously. The membrane formulations for which the theory is derived consist of a plasticized polymeric matrix that incorporates the ionophore and a mobile cation exchanger (lipophilic anion) [7,23,24]. Eqs. 2a and 2b are simplified expressions for the concentrations of the free ionophore ($C_{L,free}$) and ion-ionophore complex ($C_{IL, n+}$) as functions of space and time in a membrane with $2 \times d$ thickness. The equations are simplified by including only the terms for the side of the membrane where the species is depleted, and they also include only the first term of the summation in the equations in ref. [23], an approximation valid due to the relatively short times used in this work (i.e., $t \ll d^2/D$ where D is the diffusion coefficient in the membrane).

$$C_{L,free}(x,t) = C_{L,free}^o - \frac{kI_{app}}{nFA\sqrt{D_{L,free}}} \left[2\sqrt{\frac{t}{\pi}} \exp\left(-\frac{(d-|x|)^2}{4D_{L,free}t}\right) + \frac{d-|x|}{\sqrt{D_{L,free}}} \operatorname{erfc}\left(\frac{d-|x|}{2\sqrt{D_{L,free}t}}\right) \right] \quad (2a)$$

$$C_{IL, n+}(x,t) = C_{IL, n+}^o - \frac{I_{app}t}{nFA\sqrt{D_s}} \left[2\sqrt{\frac{t}{\pi}} \exp\left(-\frac{(d-|x|)^2}{4D_s t}\right) + \frac{d-|x|}{\sqrt{D_s}} \operatorname{erfc}\left(\frac{d-|x|}{2\sqrt{D_s t}}\right) \right] \quad (2b)$$

where $D_{L,free}$ is the diffusion coefficient of the free ionophore, I_{appl} is the constant applied current, A is the membrane surface area, F is the Faraday number, $1:k$ is the ion:ionophore

stoichiometry, n is the primary ion charge, and $D_s = \frac{(n+1)D_{R^-} - D_{IL_k^{n+}}}{n(D_{IL_k^{n+}} + D_{R^-})}$ is the effective salt diffusion coefficient in the membrane [6]. The transport number for anions is defined as

$t_- = \frac{D_{R^-}}{nD_{IL_k^{n+}} + D_{R^-}}$ and $D_{IL_k^{n+}}$ and D_{R^-} are the diffusion coefficients of the ion-ionophore complex and the lipophilic anion, respectively. $C_{L,free}^o$ and $C_{IL_k^{n+}}^o$ are the average concentrations of the free and complexed ionophore, respectively, and can easily be calculated from the total ionophore ($C_{L,tot}^o$) and lipophilic anion ($C_{R^-}^o$) concentrations added into the membrane, where

$$C_{L,free}^o = C_{L,tot}^o - \frac{k}{n} C_{R^-}^o \text{ and } C_{IL_k^{n+}}^o = \frac{C_{R^-}^o}{n}.$$

The voltage drop across the membrane has been derived previously for singly charged primary ions with 1:1 stoichiometry [7,23]. A similar equation can be derived for the general case of a primary ion charge of n^+ and ion:ionophore stoichiometry of $1:k$ as shown in Eq. 3.

$$\Delta V = \frac{RT}{nF} \ln \left(\left(\frac{C_{L,free}(-d)}{C_{L,free}(d)} \right)^k \left(\frac{C_{IL_k^{n+}}(d)}{C_{IL_k^{n+}}(-d)} \right)^{\lambda} \frac{\tilde{C}_{n^+}(-d)}{\tilde{C}_{n^+}(d)} \right) - \frac{I_{appl}}{A} R_{ohm} \quad (3)$$

where R , T , and F have their usual meanings, tildes mark solution concentrations, the parameter $\lambda = 1$ for membranes without migration (i.e., with a background electrolyte, such as ETH 500, in the membrane) and $\lambda = (n+1) \times t_-$ for membranes with migration (i.e., without a background electrolyte in the membrane), and R_{ohm} (Ωcm^2) is the area specific resistance of the membrane:

$$R_{ohm} = \frac{RT}{F^2} \int_{-d}^d \frac{dx}{\sum n_{ion} D_{ion} C_{ion}} \quad (4)$$

where $\sum n_{ion} D_{ion} C_{ion}$ is the sum of products for all ions in the membrane.

At sufficiently large applied currents the second term in Eqs. 2a and 2b will approach $C_{L,free}^o$ and $C_{IL_k^{n+}}^o$, respectively. Thus, the boundary concentration of the free ionophore, the ion-ionophore complex, or both are predicted to reach zero at some finite time called the transition or breakpoint time [3,6,7]. Eqs. 3 and 4 predict that at this time the voltage drop will be infinite. In reality, the change in the voltage drop remains finite due to violation of the model assumptions, which is discussed below. From Eqs. 2a and 2b, the expressions for the chronopotentiometric breakpoint times $\tau_{L,free}$ and $\tau_{IL_k^{n+}}$, related to the concentration polarization of the free ionophore and ion-ionophore complex, respectively, have been derived previously both for singly charged ions with 1:1 stoichiometry [7] and for multiple charged ions with $1:k$ ion:ionophore complex stoichiometry [3,6]. The equation for the free ionophore breakpoint time is:

$$\tau_{L,free}^{1/2} = \frac{nFA C_{L,free}^o \sqrt{D_{L,free}} \pi}{2 k I_{appl}} \quad (5)$$

The equation for the ion-ionophore complex breakpoint time is more complicated due to the effects of migration on the concentration profiles of charged species in current-polarized membranes:

$$\tau_{\text{II}_k^{\text{n}+}}^{1/2} = \frac{nAFC_{\text{II}_k^{\text{n}+}}^0 \sqrt{D_s \pi}}{2I_{\text{appl}} t_-} = \frac{nAFC_{\text{II}_k^{\text{n}+}}^0}{2I_{\text{appl}}} \sqrt{\frac{(n+1)D_{\text{II}_k^{\text{n}+}}(nD_{\text{II}_k^{\text{n}+}} + D_{\text{R}^-})\pi}{nD_{\text{R}^-}}} \quad (6)$$

The effect of migration can be minimized or eliminated by loading a high concentration of background electrolyte into the membrane. Accordingly, the expression for $\tau_{\text{II}_k^{\text{n}+}}$ becomes simpler for membranes with incorporated background electrolyte, as shown in Eq. 7.

$$\tau_{\text{II}_k^{\text{n}+}}^{1/2} = \frac{nAFC_{\text{II}_k^{\text{n}+}}^0 \sqrt{D_{\text{II}_k^{\text{n}+}} \pi}}{2I_{\text{appl}}} \quad (7)$$

The expression for $\tau_{\text{L,free}}$ (Eq. 5) remains the same in membranes with background electrolyte. Eqs. 5 and 6 can be used to formulate a criterion for conditions when the breakpoint related to the free ionophore concentration polarization occurs first in membranes with migration [3]:

$$\frac{C_{\text{L,free}}^0}{k C_{\text{II}_k^{\text{n}+}}^0} < \sqrt{\frac{(n+1) D_{\text{II}_k^{\text{n}+}} (n D_{\text{II}_k^{\text{n}+}} + D_{\text{R}^-})}{n D_{\text{L,free}} D_{\text{R}^-}}} \quad (8)$$

and without migration:

$$\frac{C_{\text{L,free}}^0}{k C_{\text{II}_k^{\text{n}+}}^0} < \sqrt{\frac{D_{\text{II}_k^{\text{n}+}}}{D_{\text{L,free}}}} \quad (9)$$

The equations for the ion-ionophore complex breakpoint were derived with the assumption that ion-pair formation between the ion-ionophore complex and the mobile anionic ion-exchanges sites inside the membrane is negligible [24].

MATERIALS/METHODS

Chemicals

Poly(vinyl chloride) high molecular weight (PVC), bis(2-ethylhexyl) sebacate (DOS), 2-nitrophenyl octyl ether (*o*NPOE), tetradodecyl ammonium-tertakis(4-chlorophenyl) borate (ETH 500), Calcium ionophore IV: N,N-dicyclohexyl-N',N''-dioctadecyl-3-oxapentanediamide (ETH 5234), and the H⁺-selective Chromoionophore I: 9-(Diethylamino)-5-octadecanoylimino-5H-benzo[20]phenoxazine (ETH 5294) were all Selectophore grade from Fluka. Sodium tetrakis[3,5-bis(trifluoromethyl) phenyl] borate (NaTFPB) was from Dojindo Laboratories. Tetrahydrofuran (THF) from Sigma served as the solvent for membrane casting, and polyurethane (Tecoflex SG85A, Thermedics Polymer Products, Woburn, MA) was used for the fabrication of spacer rings

Imaging of concentration profiles during chronopotentiometric experiments

The spectro-chronopotentiometric experiments were performed with a Gamry Reference 600 potentiostat/galvanostat using a spectroelectrochemical microscopy (SpECM) cell, which allowed the imaging of the concentration profiles of the free and complexed ionophores in the current-polarized membranes during the chronopotentiometric measurements [7–10]. SpECM experiments and ion-selective membrane preparations were performed as described previously [9] using a Nikon Eclipse E600 microscope (Southern Micro Instruments, Atlanta, GA) connected to a PARISS[®] (LightForm, Inc., Hillsborough, NJ, <http://www.lightforminc.com>) spectroscopic imaging spectrometer. Briefly, 240 complete spectra from 400 to 800 nm were collected across the membrane cross-section, with a spatial resolution of 1.7 μm .

The imaged membranes contained a 1:2 ratio of PVC and DOS, 2.4 mM ionophore ETH 5294, and 1.0 to 1.1 mM NaTFPB as lipophilic cation-exchanger. The membrane components dissolved in THF were poured in glass rings of 4 cm inner diameter. Membrane strips having a width ($2 \times d$) between 306 and 323 μm , and a thickness (optical pathlength) of about 190 μm were glued between the bisected parts of the spacer rings and mounted in the thin layer spectroelectrochemical cell [9]. As spacer rings, polyurethane membranes were prepared by casting 283 mg of a polyurethane/*o*NPOE mixture (41 wt. % *o*NPOE and 59 wt. % of Tecoflex SG85A) dissolved in 2 mL THF into a 30 mm i.d. glass cylinder fixed on a glass substrate. After evaporation of the solvent, the polyurethane membrane thicknesses were approximately 175 μm . The membrane separated two compartments containing deionized water with 1 mM HCl.

In the chronopotentiometric experiments, a 50 nA amplitude current pulse was applied first with a certain pulse duration. To restore the membrane concentration profiles to the equilibrium concentration profiles more quickly, a recently proposed reverse current restoration protocol was used [25,26]. For this protocol, the forward current pulse was followed by a reverse current pulse with an amplitude of -50 nA and 85–90% of the pulse duration of the initial pulse. The reverse current pulse was followed by a zero current relaxation period until the end of the experiment. Spectra across the membrane cross section were collected during the forward, reverse, and zero current steps to ensure the concentration profiles returned approximately to their original flat profiles, which usually took about 25 min. Previous work indicated that the optimal reverse current pulse should contain about 95% of the charge of the forward current pulse for experiments in which the breakpoint is not reached [25,26]. However, since one or more breakpoints were reached during the experiments in this paper, the optimal reverse current pulse contained 85–90% of the charge of the forward pulse.

The absorbance profiles across the membrane cross section were converted to concentration profiles as described previously [7], i.e., the concentrations of the unprotonated and protonated chromoionophores were calculated across the membrane cross section by using absorbance values measured at 502 and 660 nm wavelengths in combination with the corresponding molar extinction coefficients, respectively. Finally, Eqs. 1 and 2 were fitted to the concentration profiles as described previously [7,23].

Chronopotentiometric experiments with calcium-selective membranes

The chronopotentiometric experiments were performed with Ca^{2+} selective membranes in a custom-made electrochemical transport cell [6]. The effective membrane surface area in the transport cell was 0.785 cm^2 . The membrane separated two compartments filled with 20 mL of 1 mM CaCl_2 solutions. The CP current was set to 4 μA and the membrane potential was recorded every 0.5 s using Ag/AgCl reference electrodes immersed in the CaCl_2 solutions on the two sides of the membrane. The calcium-selective membranes were composed of a 1:2

ratio of PVC and DOS, with 10.4 mM ETH 5234 and either 32 or 48 mol% NaTFPB relative to the ionophore.

RESULTS/DISCUSSION

In CP experiments the applied current determines the shape of the CP transients. When the applied current is selected properly, a breakpoint (significant change in the slope) appears in the CP transients. This breakpoint in the voltage transients was shown to be caused by the free ionophore concentration approaching zero at the positive phase boundary of the membrane. Recently, we showed that under certain experimental conditions the ion-ionophore complex concentration can also approach zero [7]. This event also prompts a change in slope of the CP transients.

In Fig. 1, we show the changing absorbance profiles of the free and complexed chromoionophore ETH 5294 across a pH sensitive membrane during a chronopotentiometric experiment when the membrane is polarized with a current of 50 nA ($\sim 3.5 \mu\text{A}/\text{cm}^2$). The concentration profiles in chronopotentiometric experiments, determined from the respective absorbance profiles, are expected to show point symmetry [23]. Apparently, in the experiment shown in Fig. 1, this point symmetry is gradually lost with time. As shown in Fig. 1b, at longer times the ion-ionophore concentration is decreasing on the positive side of the membrane instead of an expected increase of the same magnitude as the decrease in the ion-ionophore concentration on the negative side of the membrane. This is a clear indication that the membrane lost most of its selectivity at its positively polarized side. The traces representing the free ionophore concentration on same side of the membrane (Fig. 1a) provide additional support for this statement. Apparently, after approximately 1 minute polarization with the 50 nA current, the free ionophore concentration is zero at the membrane solution interface. In the absence of free ionophores, the ionophore-induced selectivity of the membrane for primary ions is lost, and only limited discrimination is retained between the solution cations due to the extraction and ion-exchange properties of the membrane. However, in our experiments no interfering cations were present in the solution except for traces from impurities or contaminations. Therefore, predominately primary ions are still expected to enter the membrane even after ionophore-induced selectivity is lost. In the membrane region where the free ionophore is depleted, the ion-ionophore complex concentration begins to decrease because the migration-controlled flux of the ion-ionophore complex into the membrane is greater than the sum of the diffusion-controlled fluxes of the ion-ionophore complex and the free ionophore toward the boundary.

The membrane concentrations of the free ionophore and ion-ionophore complex at the two interfaces cannot be determined exactly from SpECM images, due to considerably light scattering. Therefore, they were estimated by fitting Eqs. 2a and 2b to the experimentally recorded concentration profiles and the fitted functions were extrapolated to the estimated coordinates of the interface, as shown in Fig. 2. In this way, the time at which the boundary concentration approaches zero can be estimated. Since the assumptions in the theory are valid only for times before the breakpoint (i.e., while the membrane selectivity is maintained), only concentration profiles that were recorded before the breakpoint time were used in the fittings. Unfortunately, the light scattering at the membrane-solution interfaces prevents also the exact demarcation of the membrane boundaries. In this work, the highest absorbances (marked by solid vertical lines in Fig. 1) resulting from the light scattering at the membrane|solution interfaces were considered as the best estimates of the boundary locations.

For example, in Fig. 2a, the free ionophore concentration appears to approach zero at the boundary just after the concentration profiles fitted for 30 s, so a breakpoint should occur around this time, assuming that the membrane boundary is located as demarked by solid vertical

line. Similarly, from Fig. 2b, the concentration of the protonated chromoionophore (ion-ionophore complex) appears to approach zero concentration just after 60 s. It should be noted, however, that quite different breakpoint times would be predicted if the membrane boundaries were assumed to be at the inner or outer dotted vertical lines. Still, the free ionophore and the ion-ionophore complex concentrations appear to approach zero at different times. Therefore, two breakpoints in the voltage-time transient are expected within the time frame of the experiment.

As forecasted by Fig. 2, two breakpoints indeed appeared in the voltage-time transient shown in Fig. 3a (marked with the vertical dashed lines). The times at which the ionophore or the ion-ionophore complex concentrations approach zero at the two phase boundaries (the zero concentration times calculated from SpECM) were compared with the breakpoint times determined by the linear extrapolation of sections on the CP transients before and after the breakpoint, as shown in Fig. 3a. To determine the zero concentration times, Eqs. 2a and 2b were used to simulate the boundary concentration-time transients when assuming different phase boundary locations, as described below. In Fig. 3, the voltage and boundary concentrations were plotted against $t^{1/2}$ rather than t because the boundary concentrations (and therefore initially the voltage) change linearly vs. $t^{1/2}$, enabling a more accurate graphical determination of the breakpoint time.

As evident from Fig. 3 and Table 1, the agreement between the calculated breakpoint times and zero concentration times is not perfect. The first obvious, source of error to test regards the contingent uncertainty in the location of the phase boundary. The calculated boundary concentration transients for different assumed boundary locations re shown in Fig. 3b. Although the location of the phase boundary has strong influence on the membrane boundary concentrations, it is unlikely that they alone cause the discrepancy between the breakpoint times and the zero concentrations times. The match between these times for the free ionophore is best when the innermost membrane boundaries are assumed to be the actual boundaries, but the match for the ion-ionophore complex is best when the outermost membrane boundaries are assumed to be the actual boundaries. However, it has to be noted that these are opposite phase boundaries and consequently an inaccurate determination of the center of the membrane could contribute to this discrepancy. But, unlike the boundaries, the center of the membrane can be determined relatively accurately by fitting the concentration profiles.

In general, for experiments performed with a variety of ETH 5294 membranes, the breakpoint related to the free ionophore concentration emerged in the CP transients before the free ionophore concentration approached zero at the phase boundary, and the breakpoint related to the ion-ionophore complex concentration emerged much later than the ion-ionophore concentration approached zero at the other phase boundary. Indeed, the breakpoint time determined from the CP transients was up to 2.5 times longer than the time when the ion-ionophore complex concentration approached zero in the SpECM recorded concentration profiles. The trends are the same when the breakpoint related to the ion-ionophore complex concentration precedes the breakpoint related to the free ionophore concentration (results not shown), i.e., the breakpoint times determined from the CP transients were much longer than the zero concentration times determined from the SpECM concentration profiles for the ion-ionophore complex.

The discrepancy between the breakpoint times and the zero concentration times could be related to the method of determining the breakpoint times in the voltage-time transients. According to Eqs. 3 and 4, the voltage should change infinitely when the concentration of the free or complexed ionophore reaches zero. However, experimentally the voltage only changes a finite amount due to violations of the theoretical assumptions.

When the free ionophore concentration approaches zero, the membrane loses most of its selectivity and solution cations will enter the membrane according to their activities in the solution and the residual selectivity of the membrane. Since in this work the solutions contain virtually no interfering cations, uncomplexed primary ions enter the membrane and their concentration increases at the membrane side of the phase boundary with the square root of time. As these “free” ions move toward the other side of the membrane, they encounter free ionophores and form complexes. Therefore, the free ionophore is gradually depleted into the bulk of the membrane, as observed in Fig. 1a. These events lead to the fast changing voltage around the time the free ionophore concentration approaches zero and the much more slowly changing voltage at longer times, as observed in Fig. 3a. Since the largest increase in voltage should occur around the time the free ionophore concentration approaches zero, it is possible that the inflection point (the time of the largest slope) may be a better estimate for the zero concentration time. In fact, Fig. 3b shows that for this membrane the inflection point (marked by a solid vertical line) occurs closer to the time predicted by the SpECM concentration profiles. However, further studies would need to be performed to determine whether it is generally more accurate. In any case, the difference between the inflection point and breakpoint times is less than 25%, which may be less than the uncertainty of the technique.

The processes are even more complex when the breakpoints are determined by the concentration polarization of the ion-ionophore complex. Consequently, to point to an exact time instant in the voltage transients when the ion-ionophore complex concentration approaches zero is even more difficult. Several processes may occur at the breakpoint time, depending on the membrane and solution compositions:

- i. In membranes that contain fixed ionic sites, like the plasticized PVC membranes used in this study with ~ 0.06 mM [27,28] fixed anionic site concentration, the concentration of the ion-ionophore complex cannot drop to zero regardless of the applied current. Instead, it approaches the fixed site concentration and then is depleted towards the membrane bulk. For membranes without a background electrolyte, this depletion of ions generates a high-resistance area with continuously increasing resistance. Thus, the measured voltage also increases continuously without a clear inflection point, as seen after the second breakpoint in Fig. 3a. The depletion of primary ions at large currents was recently modeled using the finite element method [29]. The finite element model predicted that primary ions are depleted into the bulk of the membrane in a way similar to that seen experimentally in Fig. 1b. Because the voltage increase after the breakpoint is often not completely linear, it introduces uncertainty in the determination of the breakpoint by graphical extrapolation. Finite element simulation of the processes [29] could improve the prediction of the zero concentration times from the voltage transients.
- ii. In alternative experimental setups, anions from the aqueous phase may enter the membrane at its cathodic side and contribute to the overall current. Although the function determining the level of anion coextraction is rather complex, the shape of the voltage transients after the ion-ionophore complex breakpoint can hint at the possibility of anion coextraction. In contrast to the continuous voltage increase after the breakpoint (e.g., Fig. 3a), a clear inflection point occurs in the voltage transient after the ion-ionophore breakpoint when solution anions are entering into the membrane (similar to voltage transient after the free ionophore breakpoint). Anion coextraction can also be deduced from the SpECM measurements even though anions entering the membrane have no absorbance in the visible spectral range. A continuous increase in the free ionophore concentration on the negative side of the membrane after the ion-ionophore complex concentration approaches zero indicates that very few solution anions enter the membrane.

- iii. Solution anions can also enter membranes containing a lipophilic background electrolyte, since in these membranes the flux of ion-ionophore complex due to migration is much smaller. Therefore, when the ion-ionophore complex concentration reaches the fixed site concentration, it cannot continue to be carried across the depleted layer by migration, as described in process (i). Consequently, the current must be carried by solution anions entering the membrane. Therefore, two inflection points (one for the free ionophore and one for the ion-ionophore complex) can be observed in the voltage transients of these membranes (results not shown).
- iv. In addition to the breakpoints for the free ionophore and ion-ionophore complex, voltage transients for membranes containing a background electrolyte eventually have a rapid exponential increase in voltage when a large current is applied for a long time (e.g., 9 μ A for >60 min, results not shown). Such large voltage changes were observed during coulometric ion-generation experiments when a large current was applied for a long time to membranes containing a background electrolyte [20,21]. This large voltage change was attributed to a large increase in the resistance of the membrane related to the depletion of “fixed” sites.

Conditions for a Second Breakpoint

To confirm the validity of the proposed theory for systems with complex stoichiometry, we studied the CP transients of the ETH 5234 ionophore-based calcium-selective membranes cast with 32 mol% and 48 mol% NaTFPB as a cation exchanger. ETH 5234 forms 3:1 complexes with Ca^{2+} inside the membrane, i.e., $k=3$ and $n=2$ in the corresponding equations [30,31]. In Fig. 4, we show the CP transients of these membranes as the voltage versus the square root of time and as the voltage-time gradient vs. the square root of time. Two breakpoints can clearly be discerned in the voltage curve recorded with the membrane containing 48 mol% NaTFPB, but only one is clearly discernible in the curve recorded with the membrane containing 32 mol % NaTFPB.

The breakpoints can be assigned to the ionophore or the ion-ionophore complex by using Eqs. 5 and 6 if the parameters of the equation are known (as in our case). Diffusion coefficients have been calculated previously for free and complexed ETH5234 from chronoamperometric ($D_L=2.3 \times 10^{-8} \text{ cm}^2/\text{s}$) and potentiometric ion breakthrough experiments

($D_{\text{II}_k^{n+}}=0.85 \times 10^{-8} \text{ cm}^2/\text{s}$), respectively [3]. The TFPB⁻ anion diffusion coefficient inferred from SpECM measurements is $D_{\text{R}^-} = 1.1 \times 10^{-8} \text{ cm}^2/\text{s}$ [24]. By using these diffusion coefficients in combination with Eqs. 5 and 6, one can calculate the expected breakpoint times for the membrane cast with 48 mol% NaTFPB as 5.0 $\text{s}^{1/2}$ and 12.5 $\text{s}^{1/2}$ for the free and complexed ionophore, respectively. These times compare closely with the breakpoint times identified in the CP transients of the membrane with 48 mol% NaTPFB in Fig. 4 (5.7 $\text{s}^{1/2}$ and 11.0 $\text{s}^{1/2}$ for the free and complexed ionophore, respectively). Therefore, the first breakpoint in the CP transient for this membrane belongs to the free ionophore while the second to the ion-ionophore complex. Similarly, using Eqs. 5 and 6, the breakpoint times for the membrane with 32 mol% NaTFPB can be calculated as 9.2 $\text{s}^{1/2}$ and 8.4 $\text{s}^{1/2}$ for the free and complexed ionophore, respectively. However, only one breakpoint can be identified in the CP transient of this membrane. This breakpoint appears to occur at $\sim 8.4 \text{ s}^{1/2}$, so it is most likely related to concentration polarization of the ion-ionophore complex. However, because the voltage increases continuously after this breakpoint, it conceals the breakpoint related to the concentration polarization of the free ionophore. Nevertheless, in the voltage-time gradient plot in Fig. 4 a small peak emerges at $\sim 9.5 \text{ s}^{1/2}$, which may be due to the free ionophore.

For membranes containing unknown concentrations or with unknown diffusion coefficients of species, the shape of the CP transient can indicate to which species the breakpoints belong. For

example, clear inflection points after the first breakpoint (as in Fig. 3a and in the red curve in Fig. 4) indicate that they are likely related to the concentration polarization of the free ionophore. On the other hand, the continuous increase of the voltage after the second breakpoint indicates that they are related to the concentration polarization of the ion-ionophore complex. As discussed above, if the ionionophore complex related breakpoint emerges first in the CP curves of membranes prepared without a background electrolyte (as in the black curve in Fig. 4), then the voltage continuously increases after this breakpoint and conceals the breakpoint related to the concentration polarization of the free ionophore in the membrane.

In conclusion, two breakpoints can be identified in the voltage transients of most membranes if (i) the free ionophore concentration approaches zero significantly before the ion-ionophore complex for membranes prepared without a background electrolyte, or (ii) the breakpoint times are sufficiently separated with membranes prepared with background electrolyte. In addition, the solution on the positive side must contain only primary ions.

CONCLUSIONS

In this work, chronopotentiometric (CP) transients with two transition times (breakpoints) have been presented for the first time in neutral ionophore-based cation selective membranes. Both breakpoint times can be clearly assessed in the CP transients of typical membrane formulations when the breakpoint related to the free ionophore precedes the breakpoint related to the ion-ionophore complex and the time instances of the two breakpoints are sufficiently separated from each other. With the help of the theoretical interpretation of the transients, the two breakpoint times can be utilized to follow the concentration changes of the ionophore, the ion-ionophore complex, and the mobile anionic sites or calculate their diffusion coefficients. Using spectroelectrochemical microscopy, it has been shown that the breakpoints emerge in the CP curves approximately when the free ionophore and ion-ionophore complex concentrations approach zero at the opposite membrane-solution interfaces. However, the processes occurring at the membrane phase boundaries around the breakpoint times are rather complex and different for the free and complexed ionophores, and these differences are apparent in the voltage transients. Finally, the difficulties in identifying specific times in the voltage transients when the concentrations actually approach zero are discussed.

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Fig. 1.

Absorbance profiles recorded at (a) 502 nm and (b) 660 nm, indicative primarily of the free and complexed chromoionophore concentrations, respectively, across an ETH 5294 chromoionophore based membrane during a chronopotentiometric experiment. The absorbance profiles are shown for 0, 1, 2, 3, 4, 5, and 6 min following the application of 50 nA ($\sim 3.5 \mu\text{A}/\text{cm}^2$) polarization current. The directions of the concentration changes are marked with arrows. The equilibrium concentrations of the free and complexed ionophores in the membrane were 1.4 and 1.0 mM, respectively. The estimated locations are bracketed by two dashed vertical lines, which mark the uncertainty range of the membrane|solution interface locations, i.e. the sections where light scattering occur. In 1b, on the right side of the membrane,

the ion-ionophore complex concentration profiles are labeled with the times in minutes in addition to the arrow indicating the direction of time. The inset shows in a somewhat larger magnification as the ion-ionophore complex concentration near the boundary first increases (2 min) and then decreases (3–6 min) because the free ionophore becomes completely depleted near the boundary.

Fig. 2.

Experimental (solid red) and fitted (dashed black) concentration profiles of the free ionophore (a) and the ion-ionophore complex (b) at times 0, 30, 60, and 90 s after applying 50 nA ($\sim 3.5 \mu\text{A}/\text{cm}^2$) to the same membrane as in Fig. 1. Solid vertical lines indicate the locations of the peak absorbances, and the vertical dotted lines indicate the innermost and outermost possible locations of the membrane-solution interfaces (determined from the absorbance profile in Fig. 1).

Fig. 3.

(a) Chronopotentiometric voltage vs. the square root of time transient for the experiment in Fig. 2, with dashed vertical lines marking the breakpoint times of the free ionophore (black) and the ion-ionophore complex (red), and solid vertical lines marking the times the boundary concentrations approach zero estimated from the peak absorbance boundaries. (b) Calculated boundary concentrations vs. the square root of time for the free ionophore (black) and the ion-ionophore complex (red) as they approach zero concentration. The concentrations are calculated using Eqs. 2a and 2b with diffusion coefficients and current densities determined from the fitted curves in Fig. 2. They are calculated for the locations of the boundaries at the peak absorbances (solid lines), and for the innermost and outermost possible locations of the

membrane-solution boundaries (dashed lines). The possible locations were determined from the absorbance profile in Fig. 1. Dashed vertical lines indicate the breakpoint times for the free ionophore (black) and the ion-ionophore complex (red). The solid black vertical line marks the inflection point after the free ionophore breakpoint. The breakpoint times and the inflection point were calculated from the voltage transient, as shown in Fig. 3a.

Fig. 4.

Voltage and voltage-time gradient vs. the square root of time during the application of $5.1 \mu\text{A}/\text{cm}^2$ ($4 \mu\text{A}$) to membranes containing 10.4 mM ETH 5234 with 32 mol% (black) and 48 mol % (red) anionic sites. The dashed and solid vertical lines indicate the breakpoint times calculated from the inflection point for the free ionophore and the ion-ionophore complex, respectively.

Table 1

Comparison of breakpoint times (from the voltage transient Fig. 3a) and zero concentration times (from the SpECM-derived boundary concentration transients in Fig. 3b), with experimental parameters the same as in Fig. 2.

	Transition times obtained from CP voltage transients, s		Zero Concentration times from SpECM, s	
	Breakpoint	Inflection Point	Inner Limit	Highest Outer Limit absorbance
Free Ionophore	20.0	24.8	20.8	35.8 67.6
Ion-Ionophore	120	None	38.6	63.2 114