

Electronic Supplementary Information (ESI)

Gramicidin Ion Channels in Lipid Bilayer Supported on Polyelectrolyte Multilayer Films: an Electrochemical Impedance Study

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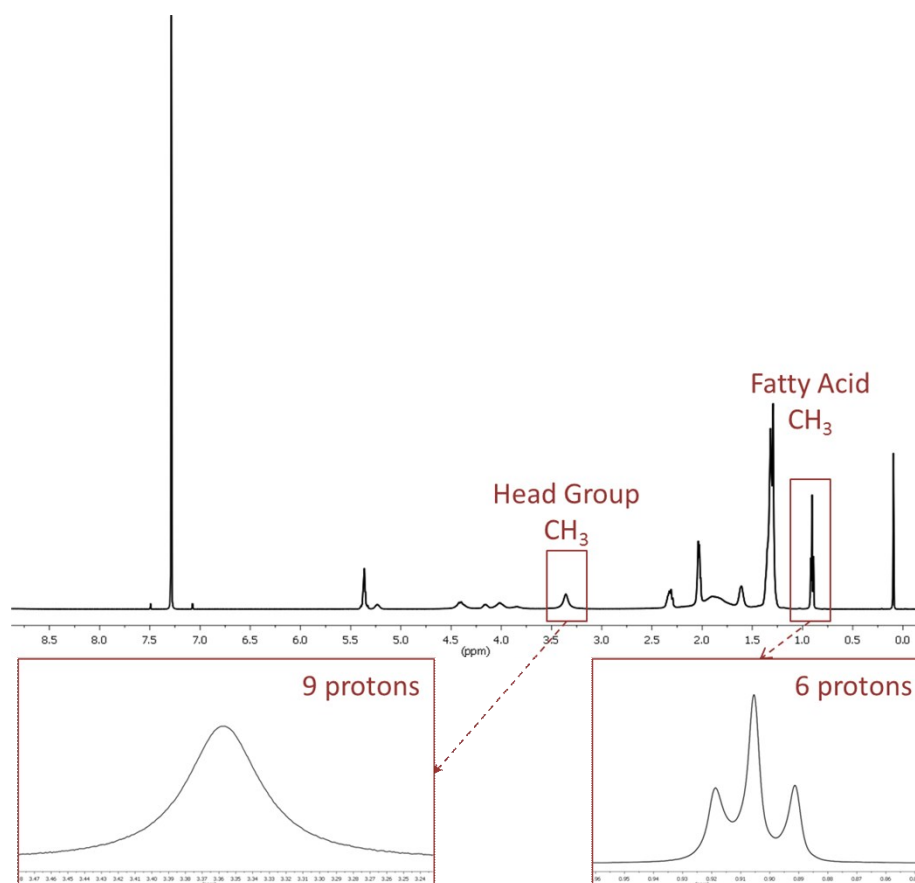


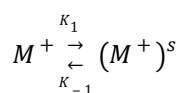
Figure S1 Typical ¹H NMR spectra of a DOPC:DOPS lipid mixture in CDCl₃. The spectral signal used to quantify the ratio of two lipids is marked in red. The lower part of the figure is a magnification of the marked peaks. The proton chemical shift at 3.3 ppm comes from the choline methyl headgroup which is only present in DOPC.

Impedance spectroscopy data were fitted to an equivalent circuit shown in the article. (see Fig. 7) This circuit resulting from the processes related to K⁺ transport through gramicidin-doped membranes consists of a parallel connection of the membrane capacitance C_m with the faradaic impedance Z . In turn, Z that can be described as a series connection of the charge transfer resistance R_1 , a parallel impedance element (C_1/R_2) representing adsorption and partitioning into the channel by the cation and the Warburg impedance element Z_w . The meanings of all the fitting parameters and how the equivalent circuit was built is shown below.

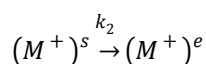
Electrochemical impedance model

A brief description of the model applied for the fitting of impedance curves is presented here. A more detailed explanation can be found in Ref [S1].

The membrane/electrolyte interface is accessed by cations, M^+ diffusion to the interface. Two consecutive steps involve ion permeation through GA-doped membrane. The first step is related to the cation adsorption at the entrance of an active GA-channel and subsequent partitioning into it. The second step involves permeation according to the single-barrier model. Formally, the reaction mechanism can be expressed as:



Equation 1



Equation 2

Where $(M^+)^s$ is the ion at the solution side of the membrane and $(M^+)^e$ is the ion at the electrode side of the membrane. the rate constants are potential (E) dependent

$$k_i = k_i^0 \exp(b_i E)$$

Equation 3

$$k_{-i} = k_{-i}^0 \exp(b_{-i} E)$$

Equation 4

Where $b_i = \alpha F/RT$ and $b_{-i} = (1 - \alpha)F/RT$, k_i^0 and k_{-i}^0 are constants independent of E and α is the transfer coefficient.

The maximum number of sites per unit surface which can be occupied by the species M^+ is characterized by a coefficient β and the fraction of sites actually occupied by θ ($0 < \theta < 1$). As a consequence the number of free electroactive sites is given by $\beta(1-\theta)$.

Mass and charge balances can be written as:

$$\beta \frac{d\theta}{dt} = k_1(1-\theta)c - k_{-1}\theta - k_2\theta \equiv g(E, c, \theta)$$

Equation 5

$$I = FA[k_1(1-\theta)c - k_{-1}\theta + k_2\theta] \equiv I(E, c, \theta)$$

Equation 6

$g(E,c,\theta)$ [Eq. (5)] and the current $I(E,c,\theta)$ [Eq. (6)] are linearized, according to Taylor series expansion retaining only terms with first-order derivatives, giving:

$$\beta \frac{d\Delta\theta}{dt} = \left(\frac{\partial g}{\partial E}\right)\Delta E + \left(\frac{\partial g}{\partial \theta}\right)\Delta\theta + \left(\frac{\partial g}{\partial c}\right)\Delta c$$

Equation 7

$$\Delta I = \left(\frac{\partial I}{\partial E}\right)\Delta E + \left(\frac{\partial I}{\partial \theta}\right)\Delta\theta + \left(\frac{\partial I}{\partial c}\right)\Delta c$$

Equation 8

Equations (7) and (8) may be obtained from equations (5) and (6) as the derivatives in stationary conditions. A small ac perturbation signal, $\Delta E = \tilde{E} \exp(j\omega t)$, is applied, and the current and concentrations oscillate around steady-state values: $I = I^{dc} + \Delta I$, $c = c^{dc} + \Delta c$ and $\theta = \theta^{dc} + \Delta\theta$, where the superscript dc indicates a steady-state term or one that does not change with the frequency of the perturbation ω , and the symbol Δ indicates a parameter oscillating periodically with time t . The resulting oscillations with time may be written as: $\Delta I = \tilde{I} \exp(j\omega t)$, $\Delta\theta = \tilde{\theta} \exp(j\omega t)$ and $\Delta c = \tilde{c} \exp(j\omega t)$.

Thus, equations (7) and (8) can be rewritten as:

$$\beta j\omega \frac{d\theta}{dE} = \left(\frac{\partial g}{\partial E}\right) + \left(\frac{\partial g}{\partial \theta}\right) \frac{\Delta\theta}{\Delta E} + \left(\frac{\partial g}{\partial c}\right) \frac{\Delta c}{\Delta I} \left(-\frac{1}{Z}\right)$$

Equation 9

$$-\frac{1}{Z} = \frac{\Delta I}{\Delta E} = \left(\frac{\partial I}{\partial E}\right) + \left(\frac{\partial I}{\partial \theta}\right) \frac{\Delta\theta}{\Delta E} + \left(\frac{\partial I}{\partial c}\right) \frac{\Delta c}{\Delta I} \left(-\frac{1}{Z}\right)$$

Equation 10

Where $Z = -\Delta E / \Delta I$ is the faradic impedance (the negative sign arises from the assumed convention in which the cathodic current is positive).

Since species M^+ diffuses towards the surface, one has

$$\frac{\Delta c}{\Delta I} = -\frac{N(\omega)}{FA}$$

Equation 11

Where $N(\omega) = \frac{1}{\sqrt{j\omega D}}$ for semi-infinite linear diffusion.

After elimination of $\Delta\theta$ from equations (9) and (10) and considering equation (11), results

$$Z = \frac{j\omega \left(\frac{\partial I}{\partial c}\right) \frac{N}{FA} j\omega \left[\left(\frac{\partial I}{\partial c}\right) \left(\frac{\partial g}{\partial \theta}\right) - \left(\frac{\partial I}{\partial \theta}\right) \left(\frac{\partial g}{\partial c}\right) \right] \frac{N}{FA} \frac{1}{\beta} - \left(\frac{\partial g}{\partial \theta}\right) \frac{1}{\beta}}{-\left(\frac{\partial I}{\partial E}\right) j\omega + \left[\frac{1}{\beta} \left(\frac{\partial I}{\partial E}\right) \left(\frac{\partial g}{\partial \theta}\right) - \frac{1}{\beta} \left(\frac{\partial I}{\partial \theta}\right) \left(\frac{\partial g}{\partial E}\right) \right]}$$

Equation 12

After evaluating the derivatives in equation (12) from equations (5) and (6) with $\alpha = 0.5$ and replacing θ by its steady state ($d\theta/dt = 0$) value the final expression for the impedance can be obtained:

$$Z = \frac{j\omega + \frac{k_1c + k_{-1} + k_2}{\beta} + \frac{k_1(k_{-1} + k_2)N}{k_1c + k_{-1} + k_2}j\omega + \frac{2k_1k_2(k_{-1} + k_2)N}{\beta(k_1c + k_{-1} + k_2)}}{\left[\frac{FA}{RT} \frac{F}{k_1c + k_{-1} + k_2} \frac{k_1(k_{-1} + k_2)}{RT} \right] j\omega + \frac{FA}{\beta} \frac{F}{RT} \frac{2k_1k_2(k_{-1} + k_2)N}{k_1c + k_{-1} + k_2}}$$

Equation 13

Finally, the impedance of the processes related to gramicidin-doped membranes consists of a parallel connection of the membrane capacitance C_m and the faradaic impedance Z .

The equivalent circuit representation of equation (13) was used to fit the experimental impedance data. The circuit components depend mainly on the values of the kinetic constants, cation concentration and temperature.

Comparison of terms of equal degree in ω in numerator and denominator of equation (13) and the expression of Z for the equivalent circuit (see article) leads to the following relations:

$$Z_w = \frac{1}{Y_0 \sqrt{j\omega}} = \frac{1}{FA} \frac{1}{\frac{F}{RT} c} N$$

Equation 14

$$R_1 = \frac{k_1c + k_{-1} + k_2}{\frac{F}{FA} k_1c(k_{-1} + k_2)}$$

Equation 15

$$R_2 = \frac{R_1(k_1c + k_{-1} + k_2)}{2 \left(\frac{k_2}{k_2} \right)}$$

Equation 16

$$C_1 = \frac{\beta FA \left(\frac{F}{RT} \right) k_1c(k_1 + k_2)}{(k_1c + k_{-1} + k_2)^2}$$

Equation 17**References**

S1. Vallejo A.E., Gervasi C.A., Impedance analysis of ion transport through gramicidin channels in supported lipid bilayers, *Bioelectrochemistry* 57 (2002) 1-7.