Phase-field modeling of electric field induced poration of lipid membranes

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Abstract

In this work, a model and simulation method to study the dynamics of pore formation and annihilation in a lipid membrane under an applied electric field has been developed. A continuum-level diffusive interface model (phase-field) method is applied to model the evolution of the pore in a lipid membrane patch. The numerical method and results are presented.

Keywords: Phase-field, Electroporation, Lipid membrane

1. Introduction

Vesicles have biomedical applications as vectors for drug and gene delivery. Manufactured vesicles can be injected with drug molecules, or other particles such as DNA, which can in theory be delivered directly to a particular region (Weaver and Chizmadzhev, 1996). An ability to release the interior of the drug in a controlled environment is crucial for any application. One possible approach is so called ‘electroporation’. Although electroporation is of interest as an alternative to viral delivery methods, it has not yet been developed to the point that it can be applied routinely in clinical practice (Smith et al., 2004). The main obstacle is the lack of a good theoretical model. Under the action of high potential difference across the biological membrane, they lose their barriers functions (electric breakdown). It is generally accepted now that electric breakdown is based on the formation of pores in the lipid areas of the membrane. It is also accepted that due to extra surface tension produced by potential difference cause the formation of these pores.

In this paper the diffusive interface approach (phase-field) is adopted in order to simulate the electroporation of a simple model system based on planar lipid bilayers. It is believed the study of planar system paves the way to study more complex systems such as lipid vesicles and biological cells.

2. Theoretical Background

2.1. Transient Pores Model

The initial observation of pores on membranes did not involve the electrical behaviour. The possibility of spontaneous poration initially suggested by two groups and later on studied more carefully (Karatekin et al., 2003; Sandre et al., 1999). The assumption is that during the constant thermal fluctuation of lipid molecules hydrophobic pores are spontaneously formed in the lipid matrix (Figure 1b). Exceeding to the critical radius, a reorientation of lipids occur and converts the pore into hydrophilic pores (Figure 1b) with the head groups forming the pore walls.

![Figure 1: Types of pores in lipid membrane](image)

The life time of the hydrophobic pores is in the order of the lipid fluctuations. Therefore, it is assumed that they are only intermediate stages in the formation of hydrophilic pores which are more stable and larger in size (Glaser et al., 1988). In this paper the intermediate stage of hydrophobic pores is neglected.

The mechanism of hydrophilic pores formation/annihilation is described by two competitive deriving force: the energy per length along the pore
edges $\gamma$, we call it line tension, and the membrane tension or the energy per area of the membrane of a flat pore-free membrane $\sigma_0$ where it is simply called the surface tension. The energy of transient pore $\Delta W_p$ is usually given for a single pore with radius of $R$:

$$\Delta W_p(R) = 2\pi \gamma R - \sigma_0 \pi R^2$$  \hspace{1cm} (1)

One can simply generalize this for pore region instead of one pore and rewrite the free energy equation in the form of two integrals. The first integral is the interracial contribution due to line tension $\gamma$ acting on the pore edges $\partial \Gamma$, and the second integral is the surface tension contribution to the porated area $\Gamma_p$ of the lipid membrane:

$$\Delta W_p = \oint_{\partial \Gamma} \gamma ds - \int_{\Gamma_p} \sigma_0 dA$$  \hspace{1cm} (2)

Surface tension $\sigma_0$ is responsible for appearance of transient pores in vesicles and lipid membranes. The surface tension for the membrane in relaxed state is very small. However, due to many reasons such as thermal fluctuations or increased hydrodynamic pressure inside the vesicle the membrane eventually becomes tense and pore formation occurs. After the pore formation and releasing the surface tension, pores will eventually reseal, the deriving force here is the line tension $\gamma$. In Figure 2 the opening and closing deriving forces are shown.

### 2.2. Electroporation Model

Many theories describing the electroporation process have surfaced in recent (and not so recent) years (Weaver and Chizmadzhev, 1996; Zimmermann et al., DeBruin and Krassowska, 1999). Electroporation is a dynamic phenomenon that depends on the local transmembrane voltage $U_m$ of the lipid part of the membrane. The induced voltage over the lipid membrane creates an extra surface tension $\sigma_e$ over the membrane in addition to the membrane tension $\sigma_0$ that already exists on the membrane. The overall surface tension then is:

$$\sigma = \sigma_0 + \sigma_e$$  \hspace{1cm} (3)

The last term is an additional surface tension due to electric potential induced over the membrane surface. Electric surface tension defined as Weaver and Chizmadzhev (1996):

$$\sigma_e = \frac{1}{2} C_{LW} \bar{U}_m^2$$  \hspace{1cm} (5)

Here $\bar{U}_m$ is the spatially averaged transmembrane voltage,

$$-\frac{U_0}{2} \quad \Phi^+ \quad \lambda^+ \quad \text{pore} \quad \Phi^- \quad \lambda^- \quad \frac{U_0}{2}$$

Here $U_m$ can be obtained from solving electric field equation of the system:

$$\nabla^2 \Phi^\pm = 0$$  \hspace{1cm} (6)
where \( E^\pm = -\nabla \Phi^\pm \) is the electric field. At the
 electrodes, the potential satisfies the boundary conditions
\( \Phi(x, y, \pm L, t) = \mp U_0/2 \), conservation of normal
currents requires
\[
C_m \frac{d\bar{U}_m}{dt} + G_m \bar{U}_m + I_p = \lambda^\pm \mathbf{n} \cdot \mathbf{E}^\pm \quad (7)
\]
where \( U_m(t) = \Phi^+(x, y, 0, t) - \Phi^-(x, y, 0, t) \) is the
membrane voltage and \( \mathbf{n} \) is the unit normal vector.
\( G_m \) is the conductance of the lipid membrane and
\( I_p \) is the current density of pore regions:
\[
I_p = \frac{\bar{U}_m}{R_p} \quad (8)
\]
where \( R_p = h/\lambda A_p \) is the pore resistance with \( A_p \)
being the area of the pore region and \( \lambda \) is the con-
ductivity of the solution filling the pore and \( h \) is the
thickness of the membrane.

At \( t = 0 \), when the field is applied, the poten-
tial is assumed to be continuous, \( \bar{U}_m(t = 0) = 0 \).
These system of equation can be solved numerically.
where \( \lambda^\pm \) is the interior and exterior electrolyte
(i.e. water) conductivity. This solution is the re-
sult of solving the electric field system of equation
and the leaky dielectric assumption applied on the
lipid membrane.

The change of the pore’s specific capacitance as
water displaces lipid to form a pore is simply
\[
C_{LW} = \left( \frac{\epsilon_w}{\epsilon_m} - 1 \right) C_m \quad (9)
\]
Here \( \epsilon_w = K_w \epsilon_0 \) is the permittivity of water and
\( \epsilon_m = K_m \epsilon_0 \) is the permittivity of lipid membrane,
and \( C_m \) is the constant capacitance per area of a
pore free membrane, i.e. \( C_m = \epsilon_m/h \) where \( h \) is the
thickness of the membrane. Typically \( K_w = 80 \)
and \( K_m = 2 \), so when the potential \( \bar{U} \) increases
\( \Delta W \) decreases which means pore nucleation is more
desirable.

3. Phase-field Formulation

We are adopting the first order phase transition
model. This model implies that broken membrane
states (pore phase) have lower free energy than in-
tact membrane states (lipid phase). This concept
first introduced on the study of stability of soap
films [Deryagin and Gutop 1962; Derjaguin and
Prokhorov 1981]. This approach assumes at the
presence of electric field the membrane breakdown
occurs because it is energetically more desirable i.e.
transitioning to more stable phase.

\[
\Delta W_p[\phi] = \int_\Gamma \gamma \left( \frac{\epsilon}{\epsilon_0} \nabla \phi \right)^2 + \frac{1}{\epsilon_0} g(\phi) \right) - \int_\Gamma \sigma (1 - H(\phi)) c_0 \quad (10)
\]
where \( g(\phi) = \frac{1}{4} \phi^2 (1 - \phi)^2 \) is a double-well potential
function and \( \epsilon \) a small length scale. The coefficient
\( \bar{\gamma} \) is related to the line tension coefficient \( \gamma \) by
\[
\bar{\gamma} = \frac{3}{4} \gamma \quad (11)
\]
The function \( H(\phi) \) is the Heaviside or the step
function. The smooth Heaviside function \( H(\phi) \approx \frac{1}{2} + \frac{1}{\cosh(k(\phi - 1/2))} \) were chosen in order to uti-
\[
\frac{\partial \phi}{\partial t} = -M \frac{\delta \Delta W_p}{\delta \phi} + \eta(x, t)
\]
\[
= -M \left( -\bar{\gamma} \epsilon \nabla^2 \phi + \frac{\bar{\gamma}}{\epsilon} \frac{dg}{d\phi} + \sigma \delta(\phi) c_0 \right) + \eta(x, t)
\]
where \( g'(\phi) = \frac{1}{2} (\phi - 3\phi^2 + 2\phi^3) \) and also the Dirac’s delta function \( \delta(\phi) = dH(\phi)/d\phi \) is the derivative of Heaviside function. \( M \) is related to the time scale for atomic rearrangement from the disordered phase to ordered one called the mobility coefficient where can be related to the diffusion coefficient \( D \) and temperature \( T \) by the Einstein relation \( D = M k_B T \), and the coefficient \( k_B = 4.11 \times 10^{-12} \) J is called is the Boltzmann’s constant, and \( \eta(x, t) \) is the stochastic noise due to thermal fluctuations and satisfies the relation:

\[
\langle \eta(x, t) \eta(\bar{x}, \bar{t}) \rangle = 2M k_B T \delta(x - \bar{x}) \delta(t - \bar{t})
\]

### 4. Numerical Method

We introduce the following non-dimensional variables:

\[
x' = \frac{x}{l_c}, \quad t' = \frac{t}{\tau}
\]

where \( l_c \) is a characteristic length of the membrane and \( \tau = l_c^2/D \) is the characteristic time associated with diffusion of the lipid molecules. Where \( D \) is the diffusion coefficient of lipid membrane and \( D = M k_B T \). Also the dimensionless surface and line tension can be acquired:

\[
\gamma' = \frac{\gamma}{k_B T}, \quad \sigma' = \frac{\sigma l_c^2}{k_B T}
\]

The non-dimensionalized and discretized form of Equation 13 is going to be:

\[
\frac{\phi^\prime_{i,j} - \phi^n_{i,j}}{\Delta t'} = \gamma' \varepsilon' \nabla^2 \phi^n_{i,j} - \frac{\gamma'}{\varepsilon'} \langle \phi^n_{i,j} \rangle - \sigma' \delta(\phi^n_{i,j}) \epsilon_0 + \eta'(x', t')
\]

The form of discrete Laplacian operator \( \nabla^2 \) is used where it introduced first by Oono and Puri:

\[
\nabla^2 \phi^n_{i,j} = \frac{\phi_{i+1,j} + \phi_{i-1,j} + \phi_{i,j+1} + \phi_{i,j-1}}{2 \Delta x'^2} - \frac{3 \phi_{i,j}}{4 \Delta x'^2}
\]

Take the lipid molecules the line tension \( \gamma \) is reported to be between 10pN to 30pN. We choose the characteristic length \( l_c = k_B T/\gamma \). This gives \( l_c = 6.4111 \times 10^{-11} \) m.

### 5. Results

![Graph showing transmembrane voltage over time for different voltages](Image)
References

DeBruin, K.A., Krassowska, W., 1999. Modeling electroporation in a single cell. I. effects of field strength and rest potential. Biophysical Journal 77, 1213 – 1224.

Derjaguin, B., Prokhorov, A., 1981. On the theory of the rupture of black films. Journal of Colloid and Interface Science 81, 108–115.

Deryagin, B., Gutop, Y.V., 1962. Theory of the breakdown (rupture) of free films. Kolloidn. Zh 24, 370–374.

Du, Q., 2011. Phase field calculus, curvature-dependent energies, and vesicle membranes. Philosophical Magazine 91, 165–181.

Elliott, C.M., Stinner, B., 2010. A surface phase field model for two-phase biological membranes. SIAM Journal on Applied Mathematics 70, 2904–2928.

Glaser, R.W., Leikin, S.L., Chernomordik, L.V., Pastushenko, V.F., Sokirko, A.I., 1988. Reversible electrical breakdown of lipid bilayers: formation and evolution of pores. Biochimica et Biophysica Acta (BBA)-Biomembranes 940, 275–287.

Karatekin, E., Sandre, O., Guittouni, H., Borghi, N., Puech, P.H., Brochard-Wyart, F., 2003. Cascades of transient pores in giant vesicles: line tension and transport. Biophysical journal 84, 1734–1749.

Sandre, O., Moreaux, L., Brochard-Wyart, F., 1999. Dynamics of transient pores in stretched vesicles. Proceedings of the National Academy of Sciences of the United States of America 96, pp. 10591–10596. URL: http://www.jstor.org/stable/48797.

Smith, K.C., Neu, J.C., Krassowska, W., 2004. Model of creation and evolution of stable electropores for dna delivery. Biophysical journal 86, 2813–2826.

Weaver, J.C., Chizmadzhev, Y., 1996. Theory of electroporation: A review. Bioelectrochemistry and Bioenergetics 41, 135 – 160.

Zimmermann, U., Pilwat, G., Riemann, F., . Dielectric breakdown of cell membranes. Biophysical Journal , 881 – 899.

Figure 5: Initiation of the pore and evolution during time ($U_0 = 1.0 \, \text{V}$)