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Deformation of Vesicles under the Influence of Strong Electric Fields II

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Dynamics of vesicle deformation in a pulsed electric field has been studied in the framework of perturbation theory. The membrane is assumed to be initially insulating and to become a conductor after a cerain period of time, t_p, by electroporation. The dynamical behavior was found to be influenced not only by t_p but also by the ratio of the conductivity inside the vesicle and that outside.

KEYWORDS: vesicle deformation, dynamics, electroporation

§1. Introduction

When a vesicle is subjected to a strong electric field, it often becomes porous, a phenomenon called electroporation.[1,2] Thus, the membrane of the vesicle can be regarded as conducting medium. In an earlier report[3] (called I hereafter), types of static deformation of vesicles having conducting membrane in static fields and dynamical behavior in pulse fields have been discussed. It was found that the deformation depends critically on the ratio of the conductivity inside the vesicle to that outside.

In I it was assumed that the electroporation occurs immediately after the field is turned on. However, in practice, the membrane may remain insulating for a significant period of time. If this is indeed the case, it is necessary to take into account the non-conducting membrane in order to describe the deformation dynamics of vesicles. Helfrich studied[4-6] the shape of a spherical vesicle having insulating lipid bilayer, but his discussion did not include the dynamical behavior. In this report, we present the results of investigations of dynamics incorporating finite periods of insulating phase of membrane.

§2. Formulation

2.1 *Electric fields and electric forces*

Two fundamental equations determining electric fields are 1) the current conservation for a stationary state, div $\boldsymbol{j}=0$, and 2) the ohm's law, $\boldsymbol{j}=\sigma\boldsymbol{E}$. These equations are solved under the following boundary conditions. The asymptotic form of the field at infinity is $\boldsymbol{E}\rightarrow E_0\hat{z}$, where E_0 is the field strength of the field applied in the z-direction. The boundary conditions on the membrane surfaces depend on whether the membrane is a conductor or an insulator.

If the membrane is an insulator, the normal component of the current at the vesicle surface must vanish. This leads to a condition that the field outside the vesicle, $\boldsymbol{E}_e$, has no normal component, $E_{en}=0$, at the surface. The field is determined uniquely by this condition.

The case of a conducting membrane has been discussed in I. It is necessary to consider the field inside the vesicle, $\boldsymbol{E}_i$, as well as $\boldsymbol{E}_e$. The boundary condition for the tangential component of the field is given as $E_{it}=E_{et}$ and that for the normal component is $\sigma_i E_{in}=\sigma_e E_{en}$.*

The electric force exerted on the vesicle surface may be calculated from the Maxwell tensor, and the normal component is

$$F_n=-\frac{\varepsilon_e}{8\pi}(E_{et})^2 \tag{1}$$

for an insulating membrane, where ε_e is the dielectric constant of the outside medium.

2.2 *Small deformation from a spherical vesicle*

The shape of a vesicle is represented in the polar coordinates by

$$r=a(1+g(\theta)), \tag{2}$$

where the deviation $g(\theta)$ is expressed in terms of the Legendre polynomials as

$$g(\theta)=\sum_{l=1}^{\infty} g_{2l}P_{2l}(\cos\theta). \tag{3}$$

The coefficients, g's, are assumed to be small and the perturbation method will be used in the following discussions.

The electric potentials ϕ_e and ϕ_i were determined in I for a conducting membrane. It can be shown that the potential ϕ_e for an insulating membrane is derived from ϕ_e for a conducting membrane by setting $\sigma_i=0$. The result is

$$\phi_e=-a_0E_0\left[\left(\frac{r}{a_0}+\frac{a_0^2}{2r^2}\right)\cos\theta + \sum_{l=1}^{\infty}\left(\frac{a_0}{r}\right)^{2l+1} C_{2l+1}P_{2l+1}(\cos\theta)\right], \tag{4}$$

where

$$C_{2l+1}=\frac{3}{2}(2l+1)\left[\frac{g_{2l}}{4l+1}-\frac{g_{2l+2}}{4l+5}\right]. \tag{5}$$

The normal component of the force acting on the vesicle can be derived from eqs. (1) and (4), and is given by

*These boundary conditions are also given by eq. (2.4) of I. However, it contains typographical errors. σ's should appear in the experssion for the boundary conditions of normal components as given herein.

$$F_n=-\frac{9}{32\pi}\varepsilon_e E_0^2\sin^2\theta\left[1-2g(\theta)+\frac{4}{3}\sum_{l=0}^{\infty}C_{2l+1}\frac{dP_{2l+1}(\cos\theta)}{d\cos\theta}\right]+O(g^2). \quad (6)$$

The leading term of this equation is identical to the expression derived by Helfrich and always produces a prolate deformation of a vesicle.

2.3 *Model of dynamics*

In I, equations of membrane motion have been constructed from the Lagrangean consisting of the inertial as well as curvature elastic energy terms. The equations also involve the electric force and the force due to the resistance of the surrounding fluid. They are reduced to linear coupled equations given as

$$\ddot{g}_2+2\Gamma\dot{g}_2+D_2g_2=2G \quad (7)$$

$$\ddot{g}_4+2\Gamma\dot{g}_4+D_4g_4=\frac{216}{35}Gg_2, \quad (8)$$

where D_2, D_4 and G are given by

$$D_2=K_2+\frac{61}{35}K_E \quad (9)$$

$$D_4=K_4+\frac{81}{77}K_E \quad (10)$$

$$G=\frac{1}{4}K_E \quad (11)$$

for an insulating membrane. Γ, K_E, K_2 and K_4 are defined in I and depend on the field strength E_0, the vesicle radius a_0 and the dielectric constant of the fluid outside the vesicle. They also contain the fluid resistance coefficient, γ, the mass factor, ν, and the curvature elastic modulus, κ. In the case of a conducting membrane, similar expressions for D_2, D_4 and G are given in I.

If the applied field is switched off, the dynamical equations are given by eqs. (8) and (9) with $D_2=K_2$, $D_4=K_4$ and $G=0$. D_2, D_4 and G for a conductiong membrane depend on the ratio $x=\sigma_i/\sigma_e$. It was shown that the driving force term, G, is positive (negative) for $x>1$ ($x<1$). The sign of G determines the mode of deformation; prolate if $G>0$ and oblate if $G<0$. In contrast, G for an insulating membrane is always positive as can be seen from eq. (11). Thus, the deformation is always prolate as pointed out by Helfrich.[6]

It should be noted from eqs. (9) and (10) that the electric field gives positive contributions to the restoring force constants. On the other hand, it was shown in I that the electric field may give negative contributions for certain values of x.

§3. Calculations

In a typical experiment, a pulsed field with the duration of t_0 is applied and the time dependence of the deformation is measured.[1] If the field is strong enough, the electro-poration occurs at $t=t_p$ ($<t_0$). After the field is switched off, the deformation continues to increase, reaching a maximum at $t=t_{max}$, and it decreases nearly exponentially to zero with the time constant t_r. The expression for t_{max} in terms of $g_2(t_0)$ and $g_2(t_0)$ is given in I. For a certain ranges of x and t_p, g_2 reaches a maximum only before the electric field is turned off and it decreases to zero monotonically thereafter. In such cases, t_{max} does not exist.

Calculations have been carried out for a typical set of values of $E_0=500$ Vcm^{-1}, $t_0=500$ μs and $a_0=10$ μm. The values of the parameters, ν and κ were the same as those used in I. The fluid resistance coefficient, γ, was set to 7.5 gcm^{-2}s^{-1} as in I. For this set of parameters, $t_r/t_0=21.5$. Figure 1 shows the time evolution of g_2 and g_4 for $x=2$. Calculations were perfomed for $t_p/t_0=0$, 0.5 and 1. The value of t_{max} was found nearly independent of t_p. The ratio t_{max}/t_0 ranges from 3.58 for $t_p/t_0=0$ to 3.45 for $t_p/t_0=1$. It can be seen that the overall behavior is essentially the same for different t_p. The maximum deformation is attained for $t_p/t_0\approx 0.5$ in this case. It can be shown that the driving force term, G, for a conducting membrane coincides with that for an inslulating membrane if $x=2/3(1+\sqrt{7})=2.43$. Since eq. (8) gives $g_2=Gt^2+O(t^3)$ for small values of t, the initial behavior is the same irrespective of whether the membrane is conducting or non-conducting. Figure 2 shows the time dependences of g_2 and g_4 for $x=2.43$ with $t_p/t_0=0$, 0.5 and 1. The difference in the time dependence for larger values of t is due to the difference between the restoring force D_2 for the conducting membrane and that for the non-conducting membrane. D_2 for a conducting membrane in a strong field can be very small and even be negative in contrast to D_2 given by eq. (9) for a non-conducting membrane.

The results of calculations for $x=0.2$ are depicted in Fig. 3. The time evolution is strongly dependent on t_p/t_0 contrary to that for $x>1$. Since the deformation of a conducting membrane is oblate for $x<1$, g_2 remains negative

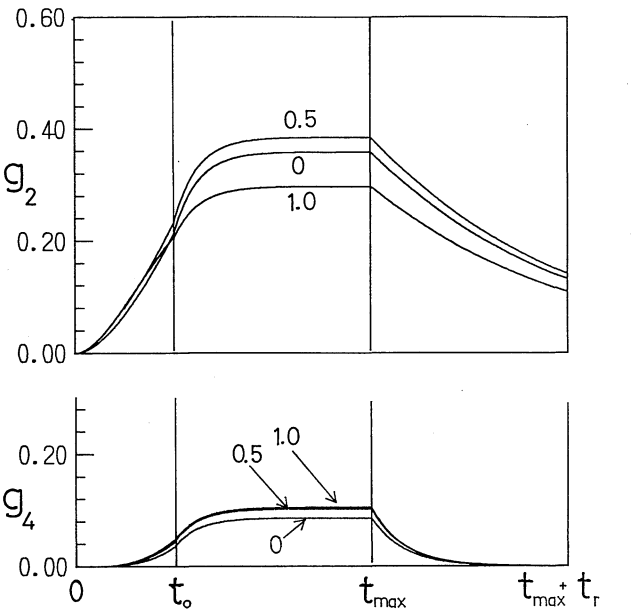

Fig. 1. Time evolution of g_2 and g_4 calculated for $x=2$. Calculations were made for $t_p/t_0=0$, 0.5 and 1. The numbers in the figure represent the values of t_p/t_0. Note that the linear scales for the regions $0<t<t_0$, $t_0<t<t_{max}$ and $t_{max}<t<t_{max}+t_r$ are different. The values of t_{max}/t_0 and t_r/t_0 are given in the text.

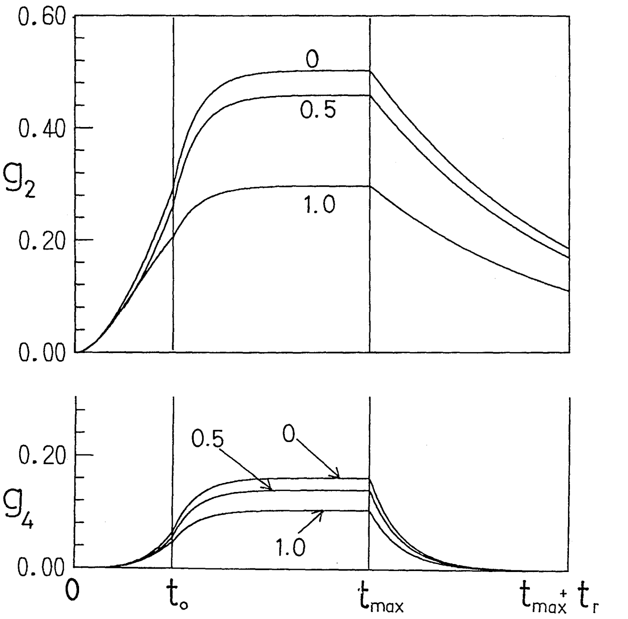

Fig. 2. Time evolution of g_2 and g_4 calculated for $x=2.43$. Calculations were made for $t_p/t_0=0$, 0.5 and 1. The numbers in the figure represent the values of t_p/t_0. Note that the linear scales for the regions $0<t<t_0$, $t_0<t<t_{max}$ and $t_{max}<t<t_{max}+t_r$ are different. The values of t_{max}/t_0 and t_r/t_0 are given in the text.

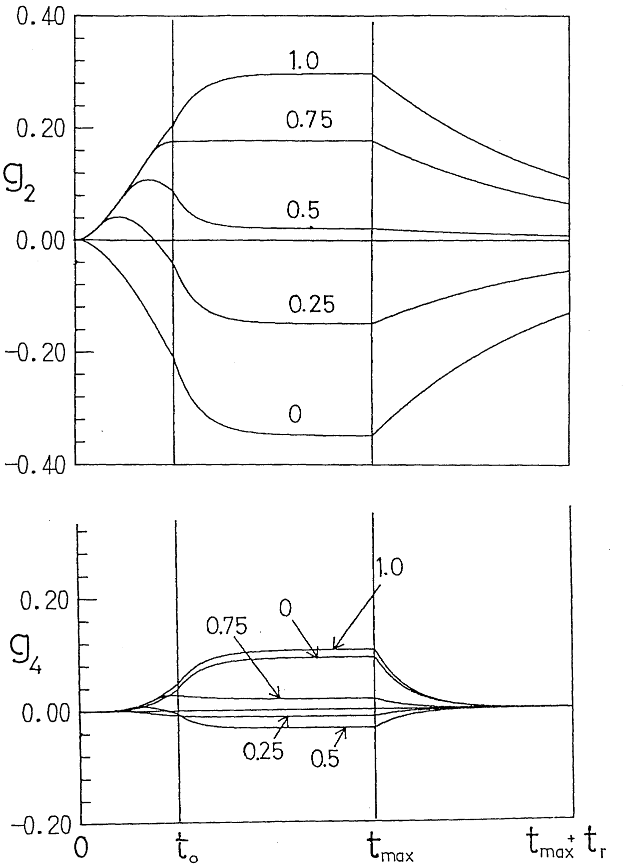

Fig. 3. Time evolution of g_2 and g_4 calculated for $x=0.2$. Calculations were made for $t_p/t_0=0$, 0.5 and 1. The numbers in the figure represent the values of t_p/t_0. Note that the linear scales for the regions $0<t<t_0$, $t_0<t<t_{max}$ and $t_{max}<t<t_{max}+t_r$ are different. The values of t_{max}/t_0 and t_r/t_0 are given in the text.

for $t_p=0$ as concluded in I. For $t_p \neq 0$, g_2 always increases initially. However, if t_p is small the dynamics is overwhelmed by that for a conducting membrane and the deformation eventually becomes oblate. For $0<t_p/t_0<0.46$, g_2 becomes eventually negative and thus has two extrema, the one at $t<t_0$ and the other at $t=t_{max}>t_0$. t_{max}/t_0 for $t_p/t_0=0$ and 0.25 are 3.56 and 3.82, respectively. For $t_p/t_0>0.74$, g_2 continues to increase after t_p and reaches a maximum at $t=t_{max}>t_0$. The ratio t_{max}/t_0 for $t_p/t_0=0.75$ and 1 are 2.02 and 3.45, respectively. For $0.46<t_0<0.74$, g_2 reaches a maximum value at a certain $t<t_0$ and then decreases monotonically to zero. In this case, t_{max} does not exist as pointed out above. In Fig. 3, the result of calculation for $t_p/t_0=0.5$ is presented with t_{max}/t_0 set arbitrarily to 4.

§4. Conclusion

In the present model the motion of the fluid surrounding the membrane is assumed to be describable in terms of the mass factor, ν, and the fluid resistance coefficient, γ. In order to go beyond this model, one needs to use the hydrodynamic equation of the fluid. However, even the simplified model used in the present calculations appears to give reasonable description of the dynamics of the vesicle deformation, and it may be useful in analyzing experimental data obtained in the field of electroporation of cells.

References

1) H. Itoh, M. Hibino, M. Shigemori, M. Koishi, A. Takahashi, T. Hayakawa and K. Kinosita, Jr.: *Proc. SPIE*, **49** (1990) 1204.
2) K. Kinosita, Jr., I. Ashikawa, N. Saita, H. Yoshimura, H. Itoh, K. Nagayama and A. Ikegami: Biophys. J. **53** (1988) 101.
3) H. Hyuga, K. Kinosita, Jr. and N. Wakabayashi: to be published in Jpn. J. Appl. Phys. **30** (1991) No. 5.
4) W. Helfrich: Z. Naturforsch. **28c** (1973) 693.
5) W. Helfrich: Phys. Lett. **43A** (1973) 409.
6) W. Helfrich: Z. Naturforsch. **29c** (1974) 182.