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The frequency dependence of phospholipid vesicle shapes in an external electric field

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Abstract. Experiments show that phospholipid vesicles exposed to AC electric field undergo a shape transition from prolate to oblate ellipsoidal shape when the frequency of the field is increased. In a theoretical model that has been devised to explain this phenomenon for nearly spherical vesicles, the vesicle shape is determined by the minimization of the total free energy of the vesicle. The two contributions to the total free energy are the membrane bending energy and the energy of the electric field. The model exhibits the same frequency-dependent prolate-to-oblate shape transition as observed in the experiment.

Key words: phospholipid vesicle · deformation · electric field

Introduction

Flaccid phospholipid vesicles with a given membrane area and a given vesicle volume can acquire a variety of shapes [1]. Their shape can also be affected by applying an external electric field. It has been observed that the shape of the vesicle depends on the frequency of the applied AC field, deforming it into a prolate rotational ellipsoid at low frequencies and into an oblate rotational ellipsoid at higher frequencies [2]. Prior theoretical analyses have either oversimplified the system and failed to predict the oblate shape in the high-frequency limit [3], or had to postulate a difference in the electrical properties between the vesicle interior and the surrounding medium [4], which is a reasonable assumption for a living cell, but not for a phospholipid vesicle. An extension of the theoretical model [3] is proposed here to include the explanation of the described phenomenon. Included are some computed results. For a detailed explanation of computational procedure involved, the reader is suggested to consult [3].

Materials and Methods

Experimental procedure

Vesicles of average diameter about 20 µm were prepared from commercially available 1-palmitoyl-2-oleoyl-*sn*-3-phosphatidylcholine (POPC) using the standard procedure [5]. When AC electric field (1-100 kHz, up to 200 V/cm) was applied to the aqueous solution of vesicles (distilled water or 0.1 $mol^{-1}\,l^{-1}$ sucrose solution), the electric forces which arise on the membrane/water boundary deformed the vesicle into a rotational ellipsoid. At low frequencies, the deformation was prolate (Fig. 1, top). By increasing the frequency of the applied field while leaving the other parametres unchanged, the vesicle was undergoing a shape change through a range of flaccid shapes to a distinctivelly oblate shape at high frequencies (Fig. 1, bottom). The transition frequency between the prolate and the oblate shape varied for different vesicles, but remained constant when repeated measurements on the same vesicles were performed. The observation was performed by means of a phase contrast microscope and a CCD video camera attached to it.

Theoretical model

The vesicle is modelled as a thin shell of lossy dielectric, immersed in an aqueous medium. Small deviations from spherical shape are allowed. Vesicle shape is determined by minimizing the total free energy of the vesicle under the constraints of a constant membrane area and a constant vesicle volume.

There are two contributions to the total free energy of the vesicle: the membrane bending energy and the energy of the electric field. The latter term is calculated as the work of electrical forces exerting on the membrane, required for the membrane displacement. In order to compute it, the Laplace equation for the electric potential has to be solved first, using the following boundary conditions: away from the vesicle the field is unperturbed, inside the vesicle the field is finite, on the membrane-water boundary the parallel component of the field is conserved and the normal component of the current including the displacement current is conserved. The electric field strength and finally the Maxwell stress tensor are then calculated from the expression for the electric potential. The Maxwell stress tensor, evaluated on both membrane-water boundaries in the direction perpendicular to the boundary, is the surface density of the force with which the electric field is acting on the membrane. Its scalar product with the membrane displacement, integrated over the total membrane area, amounts to the contribution of the electric field to the total free energy of the vesicle.

The calculation follows the procedure presented in [3] up to Eq. 18, when exact expression is calculated instead of employing the power series expansions.

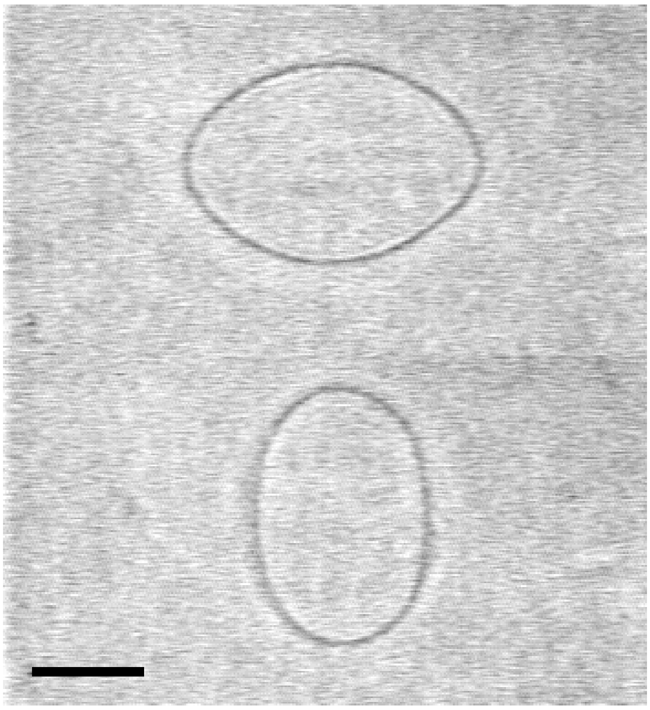

Fig. 1. A vesicle is deformed into a prolate rotational ellipsoid shape at 2 kHz (above) and into an oblate ellipsoid shape at 50 kHz (below). The bar represents 10 µm. The direction of the field is horizontal.

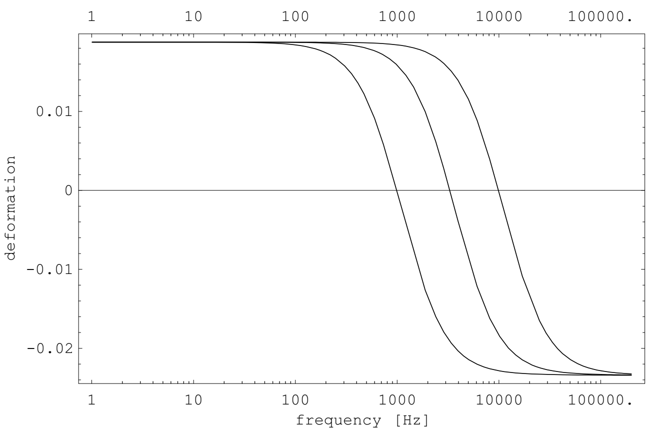

Fig. 2. Deformation expressed as a dimensionless factor of second Legendre polynomial s_2, plotted versus the frequency of the applied electric field. Positive and negative values denote prolate and oblate deformation, respectively. The three curves plotted represent three different conductivities of the aqueous medium; from left to right 3×10^{-15} S/m, 1×10^{-14} S/m and 3×10^{-14} S/m. The results are calculated for a vesicle with a radius of 4 µm and other electrical properties as found in [3].

Results

The described model extends the previous work by Winterhalter and Helfrich [3] by calculating the exact expression for the free energy instead of the power series expansion. Instead of eq. 21 in [3] we thus obtained a more complex expression for the deformation expressed as the second Legendre polynomial: $s_2 = (1/48)(\varepsilon_w r_0^4 E^2)/(k_c(1 - c_0 r_0/6))\, f(\omega)$. Here, ε_w is the dielectric constant of water, r_0 is the vesicle radius, E is the electric field strength and c_0 is the spontaneous curvature of the membrane. The expression $f(\omega)$, too long to reproduce here, is a dimensionless quantity, depending on the electrical properties of the membrane and the aqueous medium inside and outside the vesicle, the ratio between the membrane thickness and the radius of the vesicle, and the frequency of the applied field. Its dependence on the frequency of the field is shown on Fig. 2.

The predicted transition frequency for a vesicle of a given size exhibits a linear dependence on the conductivity of the aqueous medium inside and outside the vesicle. With the vesicle size increasing and other parametres kept unchanged, the transition occurs at lower frequencies. This feature promises an explanation for the experimentally observed differences in the transition frequency among different vesicles, although a systematic investigation has yet to be carried out. On the other hand, the transition frequency has proved to be largely independent on the electrical properties (i.e. conductivity and the dielectric constant) of the membrane.

The model has also proved that it is vital to account for the finite membrane thicknes, even though it is approximately three orders of magnitude smaller than the radius of the vesicle. In the limit case of infinitely thin membrane, the predicted shape at high frequencies is a sphere.

Discussion

The frequency-dependent prolate-to-oblate shape transition has been a long-neglected phenomenon. The authors of [3] have missed it because of the power series expansion they employed; in [4], the authors started with the infinitely thin membrane, and had to postulate a difference in the conductivities of the aqueous medium inside and outside the vesicle. Nevertheless, the observed shape transition does exist, and might prove itself useful for determining the conductivity in microliter samples.

References

2. Svetina S, Žekš B (1989) Membrane bending energy and shape determinition of phospholipid vesicles and red blood cells. Eur Biophys J 17:101–111
3. Peterlin P, Sevšek F, Svetina S, Žekš B (1993) Observations of giant phospholipid vesicles deformed by electric field. Acta Pharm 43:143–145
4. Winterhalter M, Helfrich W (1988) Deformation of spherical vesicles by electric field. J Coll Int Sci 122:583–586
5. Hyuga H, Kinosita K Jr., Wakabayashi N (1993) Steady-state deformation of a vesicle in alternating electric fields. Bioelectrochem Bioenerg 32:15–25
6. Needham D, Evans E (1988) Structure and mechanical properties of giant lipid (DMPC) vesicle bilayers from 20 °C below to 10 °C above the liquid crystal—crystalline phase transition at 24 °C. Biochem 27: 8261–8269