

Measurement Protocol for Planar Lipid Bilayer Viscoelastic Properties

Izidor Sabotin, Alenka Maček Lebar, Damijan Miklavčič and Peter Kramar

University of Ljubljana, Faculty of Electrical Engineering
Tržaška 25
Ljubljana, SI-1000, Slovenija

ABSTRACT

This paper describes how to estimate planar lipid bilayer's elasticity module E and surface tension σ by means of measuring its breakdown voltage and using Dimitrov's viscoelastic model of electric field-induced breakdown of lipid bilayers. Planar lipid bilayers (BLMs) were made of two components: 1-palmitoyl 2-oleoyl phosphatidylcholine (POPC) and 1-palmitoyl 2-oleoyl phosphatidylserine (POPS) in five different compositions. Folding method for forming planar lipid bilayers in the salt solution of 100 mM KCl was used. Breakdown voltages U_{br} and membrane life times t_{br} were measured by means of applying linear rising voltage signals of seven different slopes. Specific capacitances c_{BLM} of bilayers were measured with charge pulse method. Then Dimitrov's viscoelastic model was fitted to measured data allowing for estimation of elasticity module and surface tension of the lipid bilayer. Our results show that one-component bilayers composed from POPS were more stable and thus having higher breakdown voltages and elasticity moduli than bilayers composed of POPC. Surface tension values were similar regardless of the membrane composition. Values of the elasticity (E) and surface tension (σ) are comparable to those published in the literature. We conclude that the protocol used, though time consuming, is an alternative to other methods used for determination of bilayer's mechanical properties.

Index Terms — Bioelectric phenomena, biomechanics, dielectric breakdown, capacitance measurement, electric field effects, electromechanical effects, membranes.

1 INTRODUCTION

BIOLOGICAL membranes play a crucial role in living organisms. They are soft condensed matter structures that envelope the cells and their inner organelles. Biological membranes maintain relevant concentration gradients by acting as selective filters for ions and molecules. Besides their passive role, they also host a number of metabolic and biosynthetic activities [1].

Lipid bilayers are important constituents of living cell. This fact provokes a serious interest towards the study of the mechanical properties of membranes and the influence of different lipid bilayer compositions on these properties. Artificially built planar lipid bilayer is considered as a small fraction of total cell membrane, it represents the simplest model for experimental studies of membrane properties; especially because it is accessible from both sides. Latter fact makes the planar lipid bilayer especially suitable for experiments that apply electric voltage or current to the membrane and measure its response. The idea of applying voltage to lipid structures in order to determine their

properties is almost a hundred years old [2]. In 1920s physicist Fricke measured the electrical response of a red blood cell suspension as a function of frequency. He proposed a model for a biological membrane with electrically equivalent circuit comprising of a resistor and a capacitor connected in parallel. Using the equation for the parallel plate capacitor he estimated the dielectric constant (ϵ) and thickness (h) of the membrane.

The interaction of electric fields with biological membranes and pure phospholipid bilayers has been extensively studied in the last decades [3, 4]. Strong external electric field can destabilizes membranes and induce changes in their structure. The key parameter is the induced-transmembrane voltage generated by an external electric field due to the difference in the electric properties of the membrane and the external medium, known as Maxwell–Wagner polarization. According to the most widely accepted theory the lipids in the membrane are rearranged to form aqueous pores. This increases the conductivity of the membrane and its permeability to water-soluble molecules which are otherwise deprived of membrane transport mechanisms. This phenomenon is known as electroporation, sometimes referred to also as dielectric breakdown or electropermeabilization of membranes.

The stability of planar lipid bilayer is determined by the breakdown voltage which is one of the most important properties of artificial bilayers in studying electroporation. The breakdown voltage of the lipid bilayer is usually determined by a rectangular voltage pulse. The amplitude of the voltage pulse is incremented in small steps until the breakdown of the bilayer is obtained [5]. With this protocol the number of applied voltage pulses is not known in advance and each bilayer is exposed to voltage stress many times. Such a pretreatment of the lipid bilayer affects its stability and consequently the determined breakdown voltage of the lipid bilayer [6]. To avoid this inconsistency we have suggested an approach using linear rising voltage signal [7] that allows the breakdown voltage determination in a single exposure to voltage.

A macroscopic approach using the theory of elasticity of solid bodies and liquid crystals can be applied to describe mechanical properties of lipid bilayers. In 1973 Helfrich proposed a theory and possible experiments of measurement elastic properties on planar lipid bilayers [8]. As the anisotropy of lipid bilayers is clearly expressed several elasticity modules are required to describe its viscoelastic properties. Depending on the directions of the membrane deformation we distinguish volume compressibility, area compressibility, unilateral extension along membrane plane and transversal compression.

Experimentally, the material properties of bilayers have been determined using, for example, micropipette pressurization of giant bilayer vesicles [9-13] and with nuclear magnetic resonance and x-ray diffraction [14]. Analysis of the thermal fluctuations of giant vesicles using video-microscopy techniques were used by Meleard et al [15]. Lipid bilayer mechanical properties were commonly measured on giant unilamellar vesicles. Pressure was applied on a membrane with micropipette aspiration method; the properties were measured by means of video microscopy [16]. On planar lipid bilayer Winterhalter et al reports that dynamics light scattering allows to quantify viscoelastic properties in non-perturbative way. Zehl et al presented Monte Carlo simulations for investigation of the self assembly and physical properties of aggregated structures involved in a model system [17].

Transversal elasticity modulus cannot be measured directly due to small thickness of the membrane and extremely small changes of the thickness upon deformation [18]. It can be estimated through capacitance measurement with a special electrostriction method which is based on measurements of the amplitude of higher current harmonics.

It is known, that the composition of the bilayers influences its mechanical properties such as elasticity modulus and surface tension [9, 12, 18]. If the monolayer is composed of the mixture of different phospholipids, then depending on the structure of phospholipids the monolayer could be more or less densely packed. It is also known that elasticity modulus is dependent on solvent used for preparation of planar lipid bilayers and on addition of different surface active chemicals (e.g. cholesterol) [9, 18].

In our study, we measured the breakdown voltage of planar lipid bilayers of five different compositions by means of linear

rising voltage. The aim of the study was to estimate the planar lipid bilayer mechanical properties such as transversal elasticity and surface tension by means of measured planar lipid bilayer electrical properties and a viscoelastic predictive model. The main question that we addressed was how comparable are the estimated mechanical properties obtained from predictive model to reports by other researches. Influence of planar lipid bilayer composition on elasticity (E) and surface tension (σ) of planar lipid bilayer was also investigated. Proposed protocol, though time consuming, is an alternative approach to investigation of mechanical properties of planar lipid bilayers.

2 MATERIALS AND METHODS

2.1 ELECTRICAL SETUP

Our system for following up electroporation of planar lipid bilayers consists of a signal generator, a Teflon chamber and a device, which is used for measurements of transmembrane current and voltage (Figure 1). Signal generator is a voltage generator of an arbitrary type that provides voltage amplitudes from -5 V to $+5$ V. It is controlled by costume written software (Genpyrrha), specially designed for drawing the voltage signal that is used for membrane electroporation. Part of the signal generator is also a fast analogue switch. The switch disconnects the output of the signal generator and switches to the 1 M Ω resistor in 2 ns. In this way, a system discharge voltage is measured and consequently the capacitance of the lipid bilayer. Two Ag-AgCl electrodes, one on each side of the planar lipid bilayer, were immersed into the salt solution. Transmembrane voltage was measured via a LeCroy differential amplifier 1822. The same electrodes were used to measure transmembrane current. Both signals were stored in oscilloscope LeCroy Waverunner-2 354 M in Matlab format. All the signals were processed offline.

Chamber is made of Teflon. It consists of two cubed reservoirs with volume of 5.3 cm 3 each. In the hole between two reservoirs, a thin Teflon sheet with a round hole (117 μ m in diameter) was inserted. Lipid bilayer is formed by the Montal-Muller method.

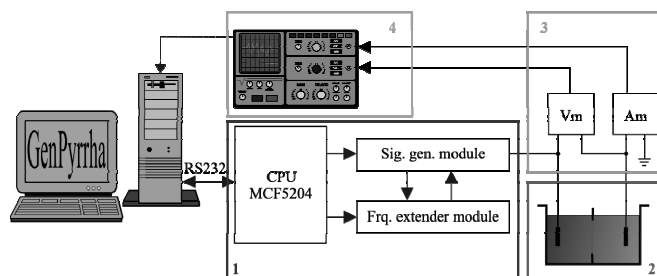


Figure 1. System for electroporation of planar lipid bilayer. 1. The microprocessor board with MCF5204 processor and two modules. One module generates arbitrary signals and the other that is realized in Xilinx, is used for frequency extension. 2. Chamber for forming lipid bilayers and two Ag-AgCl electrodes. 3. Modules for current and voltage amplification. 4. Digital oscilloscope for data storage [7].

2.2 CHEMICAL SETUP

The salt solution was prepared of 0.1M KCl and 0.01M Hepes in the same proportion. Some droplets of 1M NaOH were added to obtain pH 7.4. The lipids were prepared from POPC (1-pamitoyl 2-oleoyl phosphatidylcholine) and POPS (1-pamitoyl 2-oleoyl phosphatidylserine) in powder form (Avanti Polar-Lipids Inc. ZDA). Lipids were melted in solution of hexane and ethanol in ratio 9:1. The mixture of hexadecane and pentane in ratio 3:7 was used for torus forming. When preparing two-component bilayers both species of lipids were mixed together in appropriate ratio in small plastic tube before they were applied. Five different compositions of lipids were used. Two of them were made exclusively of one component. Two-component bilayers were made of mixture of POPC and POPS in three different ratios: 3:1, 1:1 and 1:3.

2.3 MEASUREMENT PROTOCOL

Measurement protocol consisted of two parts: capacitance measurement (Figure 2A) and lipid bilayer breakdown voltage measurement (Figure 2B). Capacitance and the breakdown voltage were determined for each lipid bilayer. The capacitance of lipid bilayer was measured with discharge method as previously described [7]. The membrane capacitance (C_{BLM}) was normalised to the surface area of the orifice for comparison with other studies (c_{BLM}).

We determined breakdown voltage (U_{br}) of the lipid bilayer by the linear rising signal. The slope of the linear rising signal (k) and the peak voltage of the signal has to be selected in advance. Seven different slopes were selected. Breakdown voltage was defined as the voltage at the moment t_{br} when sudden increase of transmembrane current was observed. Time of breakdown t_{br} was defined as a lifetime of the lipid bilayer at a chosen slope of the linear rising signal [7]

2.4 STATISTICS

To compare breakdown voltages and specific membrane capacitances of the lipid bilayers exposed to voltage signals of different slopes (k) one way ANOVA test was used. All pairwise multiple comparisons were made by Tukey's test. When statistically analyzing data of bilayers composed of POPS exclusively Kruskal-Wallis one-way analysis of

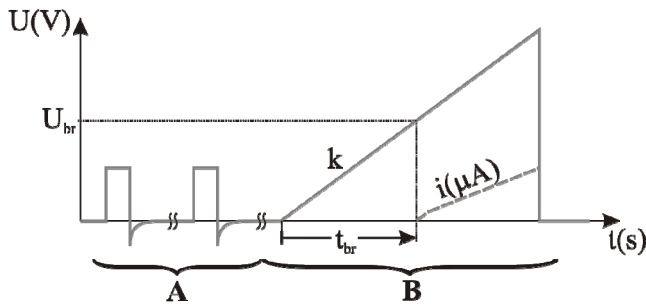


Figure 2. Measurement protocol: A) capacitance measurement of lipid bilayer was measured in two steps. In the first step we measured capacitance of the electronic system without lipid bilayer. Second step was measuring capacitance of electronic system with lipid bilayer and salt solution. B) voltage breakdown measurement with linear rising signal [7].

variance on ranks test was used, as the population did not fulfil all the requirements for one way ANOVA test i.e. the equality of variance between the compared populations. Pairwise multiple comparisons were performed by Dunn's method.

When comparing the mean breakdown voltages at the same slopes (k) between one component lipid bilayers unpaired t-test was used. As the variances of mean breakdown voltages at slopes $k=7.8$ kV/s and $k=11.5$ kV/s were statistically different, the comparisons were made with Mann-Whitney's test. We rejected the null hypothesis of analyses if the p-value of the test was less than 0.05 ($p < 0.05$) regardless of the test used.

2.5 DETERMINATION OF ELASTICITY AND SURFACE TENSION

For determination of viscoelastic properties we used the model of Dimitrov [19]. The model considers the lipid bilayer as a thin viscoelastic film with fluctuating surfaces, bounded by two semi-infinite bulk phases. The assumption that the membrane behaves as a viscoelastic, isotropic material, represented as a standard solid model, composed of a Kelvin body in series with a linear spring, is made. Originally, this model predicts the critical voltage (U_c) and critical time (t_c) needed to collapse a membrane at applied voltage. In our study critical voltage corresponds to breakdown voltage (U_{br}) and critical time to life time (t_{br}) of planar lipid bilayer. The parameters of model are Young's transversal elasticity modulus (E), surface tension (σ), viscosity (μ), thickness of the membrane (h) and permittivity of membrane (ϵ_m). If the bilayer's life time limits to infinity, the model of Dimitrov yields:

$$U_c = \sqrt[4]{\frac{8E \cdot \sigma \cdot h^3}{\epsilon_m^2 \epsilon_0^2}} \quad (1)$$

In our experimental protocol (Figure 2) planar lipid bilayer lifetime (t_{br}) is measured and can not be infinite. Therefore generic model equation that still contains (t_{br}) is more adequate [19]:

$$U_{br} = \sqrt[4]{\frac{1}{\epsilon_m^2 \epsilon_0^2} \left[8Eh^3 \sigma \left(1 + \frac{3\alpha\mu}{Et_{br}} \right) \right]} \quad (2)$$

In equation (2) we consider:

$$c = \frac{\epsilon_m \epsilon_0}{h} \quad (3)$$

$$a = \frac{8}{c^2} \sigma Eh \quad (4)$$

$$b = \frac{24}{c^2} h \sigma \alpha \mu \quad (5)$$

Where c corresponds to planar lipid bilayer capacitance normalized with the surface area c_{BLM} .

Equation (2) can be written as:

$$U_{br} = \sqrt[4]{a + \frac{b}{t_{br}}} \quad (6)$$

Using nonlinear regression, the viscoelastic model in equation 6 was fitted to the data by Generalized Nonlinear Non-analytic Chi-Square Fitting function in Matlab [20-22]. Parameters a and b , which are obtained through fitting, served to calculate Young's elasticity modulus (E) and surface tension (σ) of planar lipid bilayer according to following two equations derived from (3,4,5):

$$E = 3 \frac{a}{b} \alpha \mu \quad (7)$$

$$\sigma = \frac{bc^2}{24\alpha\mu h} \quad (8)$$

Specific capacitance (c_{BLM}) has been measured and can be used in a model. Other parameters such as thickness ($h = 3.5$ nm), viscosity ($\mu = 6$ Ns/m²) and $\alpha = 2$ were used in calculation according to the reference [19].

3 RESULTS

We measured specific capacitance (c_{BLM}), life time (t_{br}) and breakdown voltages (U_{br}) for five different planar lipid bilayer compositions. By means of viscoelastic model we estimated transversal elasticity (E) and surface tension (σ) of planar

Table 1. Fitted parameters (a) and (b), measured capacitance (c_{BLM}) and calculated Elasticity (E) and Surface tension (σ) for five different planar lipid bilayer compositions are gathered in table with their standard deviation.

Mixture	a	b	c_{BLM}	E	σ
POPC:POPS	10 ⁻² V ⁴	V ⁴ μs	μF/cm ²	N/cm ²	10 ⁻⁵ J/m ²
1:0 (n = 106)	3.20 ± 1.37	5.0 ± 0.8	0.51 ± 0.07	23.09 ± 0.37	12.90 ± 3.50
3:1 (n = 89)	2.33 ± 0.75	4.8 ± 0.6	0.17 ± 0.06	17.61 ± 0.21	1.37 ± 0.96
1:1 (n = 60)	1.99 ± 0.64	3.9 ± 0.5	0.31 ± 0.07	18.48 ± 0.24	3.69 ± 1.67
1:3 (n = 75)	4.70 ± 1.06	7.4 ± 1.5	0.24 ± 0.14	22.90 ± 0.28	4.22 ± 4.92
0:1 (n = 100)	8.36 ± 2.14	5.6 ± 1.1	0.41 ± 0.13	53.37 ± 0.65	9.41 ± 0.60

lipid bilayer. All measured data are in agreement with our previously published data [7]. Fitted parameters (a) and (b), measured capacitance c_{BLM} , and calculated elasticity (E) and surface tension (σ) data with their standard deviation are collected in Table 1. For all regression curves in Figure 3 the 68% confidence interval of the fit upper and lower border has been plotted by dot-dashed curves.

For membranes composed exclusively of POPC ($n = 106$) membranes were included and mean c_{BLM} value with standard deviation was measured to be 0.51 ± 0.07 μF/cm². U_{br} increased with increasing steepness (k) of the voltage applied as seen in Figure 3 at POPC:POPS = 1:0. Pairwise statistical comparison of the mean U_{br} were made. In general, mean U_{br}

at steeper slopes of linear rising voltage applied were statistically significantly higher from mean U_{br} at less steep voltage ramps applied. U_{br} at $k = 48.1$ kV/s was statistically significantly higher than U_{br} measured at the rest of applied slopes of voltage signal. At $k = 21.6$ kV/s U_{br} was statistically significantly higher from U_{br} at slopes $k = 11.5$ kV/s or less and at $k = 16.7$ kV/s U_{br} was statistically significantly higher from U_{br} at slopes $k = 7.8$ kV/s or less. Mean U_{br} values at slopes $k = 4.8$ kV/s, $k = 7.8$ kV/s and $k = 7.8$ kV/s showed no statistical difference. Similar statistical analysis was performed for mean breakdown voltages for all membrane compositions and similar statistical results, with respect to the voltage signal slopes were obtained irrespective of the membrane composition. From the fitted Dimitrov model function to the mean breakdown voltages we obtained the elasticity modulus E and surface tension σ for lipid membranes composed of POPC. We estimated the elasticity modulus to be 23.09 ± 0.37 N/cm² and surface tension $(12.90 \pm 3.50) \times 10^{-5}$ J/m².

For membranes composed of POPS exclusively ($n = 100$) membranes were included in defining mean c_{BLM} , t_{br} and U_{br} values. Mean specific capacitance c_{BLM} was measured to be 0.41 ± 0.13 μF/cm². A trend of increasing U_{br} with respect to steeper applied voltage slopes is seen in Figure 3 at POPC:POPS = 0:1. Between one component lipid bilayers i.e. membranes composed of POPC and POPS exclusively, statistical analysis of mean U_{br} values at same voltage slopes applied was made. Mean U_{br} values at slopes $k = 4.8$ kV/s, $k = 5.5$ kV/s, $k = 7.8$ kV/s, $k = 21.6$ kV/s, $k = 48.1$ kV/s measured on lipid bilayers composed of POPS were significantly higher from mean U_{br} values measured on membranes composed of POPC only. At slopes $k = 11.5$ kV/s and $k = 16.7$ kV/s no statistical difference was observed as the probability values for rejection of the null hypothesis were $p = 0.069$ and $p = 0.259$ respectively. The estimated elasticity modulus E for bilayers composed of POPS was 53.37 ± 0.65 N/cm² and the surface tension was evaluated to $(9.41 \pm 0.60) \times 10^{-5}$ J/m².

For membranes composed of POPC:POPS = 3:1 ($n = 89$) membranes were used in the analysis. Measured c_{BLM} was 0.17 ± 0.06 μF/cm². A trend of increasing U_{br} with respect to steeper applied voltage slopes is seen in Figure 3 at POPC:POPS = 3:1. The estimated elasticity module was 17.61 ± 0.21 N/cm² and surface tension was $(1.37 \pm 0.96) \times 10^{-5}$ J/m².

For membranes composed of POPC:POPS mixed in 1:1 ($n = 60$) membranes were included in the analysis. The mean specific capacitance c_{BLM} was 0.31 ± 0.07 μF/cm². Distinctive U_{br} dependence on the linear rising voltage applied is seen in Figure 3 at POPC:POPS = 1:1. The estimated elasticity module was 18.48 ± 0.24 N/cm² and surface tension was $(3.69 \pm 1.67) \times 10^{-5}$ J/m² respectively.

For membranes composed of POPC:POPS = 1:3 ($n = 75$) membranes were used in the analysis. Measured c_{BLM} was 0.24 ± 0.14 μF/cm². A trend of increasing U_{br} with respect to steeper applied voltage slopes is seen in Figure 3 at POPC:POPS = 1:3. We estimated elasticity E to 22.90 ± 0.28 N/cm² and surface tension σ to $(4.22 \pm 4.92) \times 10^{-5}$ J/m².

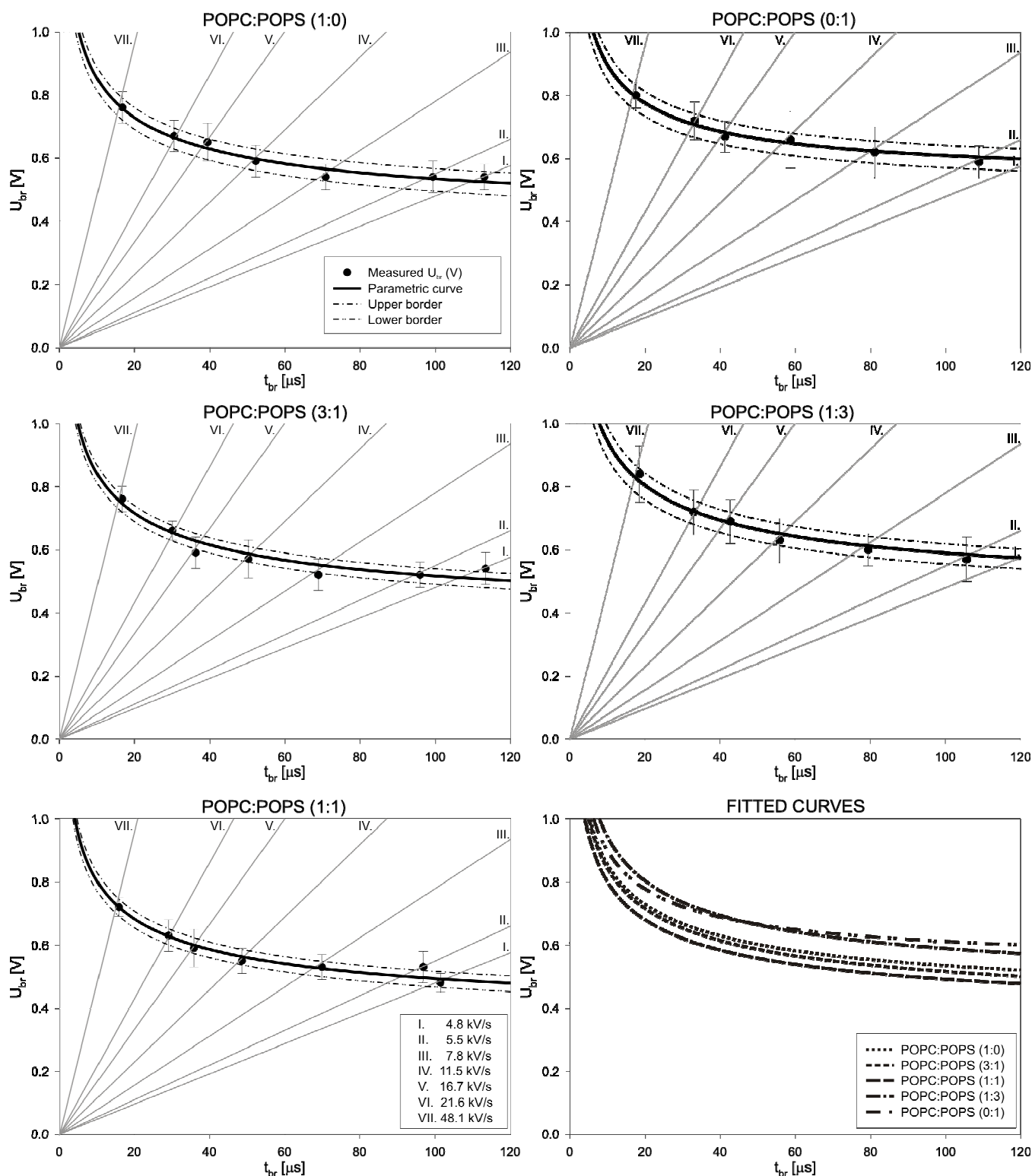


Figure 3. Dependence of the membrane breakdown voltage U_{br} and life time t_{br} on steepness of the applied linear rising voltage. All seven slopes used are denoted with roman numbers and plotted in gray color. The error bars in U_{br} measurements represent standard deviation of the data. Composition of the membrane experimented on denotes the title of the graphs meaning the volumetric ratio of the two lipids used (1-pamitoyl 2-oleoyl phosphatidylcholine POPC and 1-pamitoyl 2-oleoyl phosphatidylserine POPS). The black solid line represents the fitted Dimitrov model function on the data measured. Corresponding dash-dotted lines represent the 68% confidence interval of the fit (upper border and lower border). Fits for all compositions are plotted on the graph in bottom-right corner.

4 DISCUSSION

The breakdown voltage of planar lipid bilayers of five different compositions were measured by means of linear rising voltage. The aim of the study was to estimate the planar lipid bilayer mechanical properties such as transversal elasticity and surface tension by means of measured planar lipid bilayer electric properties and a predictive model.

Dimitrov's viscoelastic model of electric field-induced breakdown voltage of lipid bilayers predicts a dependence on electric pulse length to the electroporation thresholds of the electromechanical theory [19]. Originally it predicted a minimum critical voltage, for which the membrane becomes unstable at long pulse durations. Troiano et al. reported that this model did not fit to their measured data. The breakdown voltage of the lipid bilayer in their experiments was however determined by repeatedly applying a rectangular voltage pulse; the amplitude of the voltage pulse was incremented in small steps until the breakdown of the bilayer was obtained [5]. Using such a protocol the number of applied voltage pulses is not known in advance and each bilayer is exposed to voltage stress several and different times. Such a pretreatment of the lipid bilayer most probably affects its stability and consequently the determined breakdown voltage of the lipid bilayer [6]. By using a linear rising voltage signal, as we proposed in our previous study, we avoid this inconsistency. The breakdown of planar lipid bilayer is forced in a single voltage exposure. As a result of this protocol standard deviation of bilayer's life-time t_{br} at particular voltage slope was reduced to 10% or less. When a rectangular pulses are used, the bilayer breaks down at a random life time during the pulse. The consistency of t_{br} measurement enabled us to use generic model of Dimitrov in order to estimate bilayer's elasticity modulus (E) and surface tension (σ).

Lipid bilayers have been shown to rupture at surface tensions in a range from ~ 0.01 to $25 \cdot 10^{-3} \text{ J/m}^2$ depending on the lipid composition [9, 11, 19, 23-26]. Our results are within this interval though it has to be stressed out that uncertainty of our estimation is rather high, especially in the case of 3:1 and 1:3 mixtures. One of the reasons for that could be also the selected value of the thickness (h) which is not exactly determined for each lipid composition. On the other hand, it has been argued [27] that the electric field causes a lateral stress in the membrane that directly influences the interfacial tension and therefore has a dominant role in determining headgroup packing and pore formation.

Transversal elasticity of a planar lipid bilayer can be determined only by stressing the membrane in some way. Hianik measured the elasticity of planar lipid bilayer with electrostriction method based on measurement of the amplitude of higher current harmonics [18]. For a bilayer composed of eggPC a transversal elasticity modulus measured was on the interval from 10^5 Pa (10 N/cm^2) at lower frequencies to 10^7 Pa (10^3 N/cm^2) at higher frequencies ($\sim 2 \text{ kHz}$). He also observed the dependence of E on various solvent used. For solvents with shorter hydrocarbon chains (n-heptan, n-decan) elasticity values as mentioned above were measured, but for solvents with longer hydrocarbon chains (n-hexadecan), two orders of magnitude higher values of elasticity were reported. We used n-hexan and thus our determined elasticity moduli are comparable with Hianik's results at low frequencies using solvents with shorter hydrocarbon chains.

The bilayers were composed of two lipid species that are present in biological membranes. A lipid molecule of POPS has negatively charged headgroup while a molecule of POPC has electrostatically neutral headgroup (zwitterion). Lower breakdown voltages and consequently lower elasticity modulus and surface tension were expected for POPS bilayers due to repulsive electrostatic forces between the lipid molecules. On the contrary higher breakdown voltages for bilayers composed of POPS were measured. The elastic bending moduli of vesicles is one of the key predictors of membrane strength [28]. In our case estimated elasticity module for POPS was higher than that of POPC planar lipid bilayers. This is not only a consequence of higher breakdown voltage but also the result of other parameters of Dimitrov's viscoelastic model. Maier et al. concluded that breakdown voltage and consequently membrane stability is independent of the polarization of headgroups of lipids [29]. Similar to our results they measured the breakdown voltage for bilayers composed of POPS to be about the same, or slightly higher than breakdown voltages of POPC membranes. Similar Winterhalter et al. reports that the stability of the membrane was the same for lipids with charged (PS) and uncharged (PC) headgroups [30].

Another report that supports our findings comes from MD simulations performed by Pandit et al. [31]. They simulated a lipid bilayer composed of Dipalmitoylphosphatidylcholine (DPPC) and Dipalmitoylphosphatidylserine (DPPS). As the polar headgroups of the lipids under investigation were the same as ours we can draw parallels between their and our conclusions. They discovered that a single molecule of DPPC in the bilayers prefers complexation with two other lipids, whereas DPPS molecule prefers complexation with four. As complexation with more molecules means more stable bilayer structure, the reason why the POPS bilayers are more stable can be explained.

When two component lipid bilayers were investigated measured U_{br} and estimated E and σ were in the range of obtained values for pure POPC bilayers. This brings us to the conclusion, that the two-component bilayer's mechanical properties are predominantly determined by the least stable component – in our case this is the POPC.

5 CONCLUSION

The aim of our study was to estimate the planar lipid bilayer mechanical properties such as elasticity and surface tension by means of measured planar lipid bilayer electric properties using a viscoelastic predictive model. The protocol of the measurement of viscoelastic properties is time consuming due to the fact that planar lipid bilayer breakdown voltage and life time have to be measured at several steepnesses of linear rising signal. Nevertheless, the values of the transversal elasticity (E) and surface tension (σ) obtained with our method are comparable with previously published results in literature obtained with other methods. We also found that POPS lipids form stronger bilayers in comparison to POPC and have higher elasticity modulus. Furthermore this is reflected in two-component bilayer's mechanical properties which are dominantly determined by the least stable component.

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Izidor Sabotin was born in Murska Sobota, Slovenia in 1981. He received the B.Sc. degree from the Faculty of Electrical Engineering, University of Ljubljana, Slovenia in 2008. Currently he works as a researcher at the Faculty of Mechanical Engineering in Ljubljana. His field of research is microtechnologies and its manufacturing processes in connection with microfluidic devices and biomedical engineering, especially in the field of electroporation.



Alenka Maček Lebar was born in 1967 in Ljubljana, Slovenia. She received the M.Sc. and Ph.D. degrees in electrical engineering from the University of Ljubljana, Ljubljana, Slovenia, in 1995 and 1999, respectively. She is an Assistant Professor on the Faculty of Electrical Engineering, University of Ljubljana. Her main research is directed toward the biomedical engineering field, especially in the field of electroporation.



Damijan Miklavčič was born in Ljubljana, Slovenia, in 1963. He received the Ph.D. degree in electrical engineering from the University of Ljubljana, Ljubljana. He is currently a Professor with the Faculty of Electrical Engineering and the Head of the Laboratory of Biocybernetics, University of Ljubljana. He is active in the field of biomedical engineering. His research interest in the last years focuses on electroporation-assisted drug and gene delivery, including cancer treatment by means of electrochemotherapy, tissue oxygenation, and modeling.



Peter Kramar was born in Kranj, Slovenia in 1977. He received the B.Sc. degree from the Electrical Engineering, University of Ljubljana, Faculty of Electrical Engineering, Ljubljana, Slovenija in 2003, and the M.Sc. degree in 2005. He is a teaching assistant at University of Ljubljana, His main research area is electroporation of planar lipid bilayers.