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SPONTANEOUS OSCILLATION OF ELECTRICAL POTENTIAL ACROSS ORGANIC LIQUID MEMBRANES

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An artificial membrane was studied consisting of an oil layer, nitrobenzene containing picric acid, imposed between two aqueous phases, one of which contained 5 mM hexadecyltrimethylammonium bromide (CTAB) and 5% ethanol. It was found that this system shows rhythmic and sustained oscillation of the electrical potential within the range 150–300 mV with an interval of 2–3 min. In the absence of CTAB or ethanol, no oscillation was observed. It is indicated that in this experiment the concentrations of the solutes are far-from-equilibrium, i.e., the hydrophilic substance, picric acid, was dissolved in the organic phase and the hydrophobic substance, CTAB, was dissolved in the aqueous phase. In addition, the presence of an unstirred layer was suggested to be essential for generating such electrical oscillations.

1. Introduction

Various biological systems exhibit rhythmic phenomena, e.g., oscillation of cells of nerve and muscle tissue, oscillation in mitochondria, and biological clocks. These phenomena have recently received much attention as 'dissipative' effects caused in far-from-equilibrium concentrations [1–3]. Oscillatory phenomena have also been found in artificial membrane systems [4–8], the oscillatory variables being the electrical potential of the membrane and/or the hydrostatic pressure between the compartments on the two sides of the membrane.

Recently, we found that electrochemical oscillations can be produced even in a system consisting of oil and water phases. As an extention of such study, we subsequently investigated an artificial membrane consisting of an oil layer containing picric acid imposed between two aqueous phases, one of which contained hexadecyltrimethylammonium bromide (CTAB). The present report describes the result of our experiments, indicating that this system shows rhythmic and sustained variation of electrical potential within the range 150–300 mV between the two aqueous phases.

2. Materials and method

Experiments were performed in a U-shaped apparatus made of glass tubing (12 mm inner diameter). A solution (4 ml) of 1.5 mM picric acid in nitrobenzene was placed in the base of the U cell. Aqueous solutions (10 ml on each side) were added simultaneously to the arms of the U cell and floated on the organic phase. The solutions were allowed to stand but were not stirred. The entire cell was thermostatically regulated at 18°C throughout the measurements. The voltage across the liquid membrane was measured with a Hitachi-Horiba F-7 pH/mV meter connected by two salt bridges to two Ag/AgCl electrodes. All reagents were commercial products. Nitrobenzene was purified by distillation.

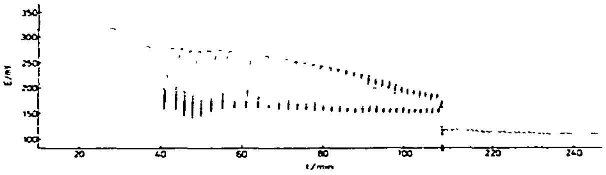

Fig. 1. Oscillations of electrical potential between the two aqueous phases. A solution (4 ml) of 1.5 mM picric acid in nitrobenzene was placed in the base of the U cell. Aqueous solutions (10 ml on each side) were added simultaneously to the arms of the U cell and floated on the organic phase. The aqueous solution in the left arm contained 5 mM CTAB and 5% (v/v) ethanol and that on the right contained 3.0 M KCl. In the recordings of the electrical potential, an upward change denotes an increase in the positive charge in the right aqueous phase.

3. Results and discussion

Oscillations of electrical potential are exemplified in Figs. 1 and 2. In both experiments, the aqueous solution contained 5 mM CTAB and 5% (v/v) ethanol in the left arm of the U cell. The aqueous solution in the right arm contained 3.0 M KCl in the experiment for fig. 1, and 0.1 M potassium phosphate buffer, pH 7, in that for fig. 2. No oscillations were observed in the absence of ethanol. Moreover, oscillations did not occur when CTAB was replaced by an anionic detergent, such as sodium hexadecylsulfonate. Fig. 1 shows that the oscillation started approx. 40 min after the organic phase came into contact with the two aqueous solutions and that it continued for approx. 4 h. The amplitude of the oscillation was approx. 150 mV initially and decreased gradually with decrease in periodicity. A similar phenomenon was observed when an aqueous solution of 3.0 M NaCl was placed in the right arm of the apparatus. On the other hand, when the aqueous solution in the right arm was replaced by potassium phosphate buffer (fig. 2), the amplitude of the oscillation became as great as approx. 300 mV though the oscillations continued for only about 15 min.

This difference in the time course of change of electrical potential is attributable to the presence of phosphate anion (P_i) in the experiment for fig. 2. P_i has been regarded as a lipophilic anion. For example, a recent study on an artificial lipid membrane showed that P_i increases the membrane permeability to hydrophobic cations (cyanine dyes), and as a result increases the conductance of the bilayer membrane [10]. It is thus possible that P_i increases the permeability of ionic species through the liquid membrane and that this causes the difference in the oscillatioins shown in figs. 1 and 2. It is noteworthy that the electrical potential decays towards 0 mV faster in fig. 2 than in fig. 1. This may again be due to the effect of P_i in increasing the membrane permeability. From these experimental results, it is speculated that the oscillations of electrical potential are generated by periodic movements of ionic species across the organic phase, probably forming 'inverted micelles' consisting of hexadecyltrimethylammonium cations and counter anions.

Recent, we found that sustained, regular oscillation of the electrical potential occurred in a two-phase system of a solution of 1.25 mM picric acid in 2-nitropropane and an aqueous solution of 5 mM CTAB [9]. It was found that when the

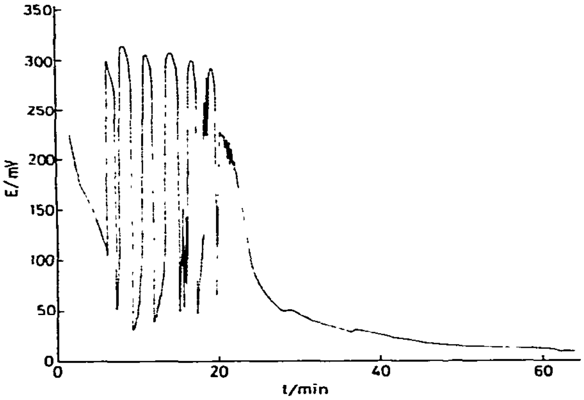

Fig. 2. Variation of electrical potential between the two aqueous phases. Experimental conditions were as for fig. 1 except that 0.1 M potassium phosphate buffer (pH 7) was used in the right aqueous phase instead of an aqueous solution of KCl.

solutions were not stirred, the oscillations were constant in period (0.5–2 min) and continued for 1–2 h, but that no oscillations occurred when the solutions were stirred. The amplitude of the electrical oscillation was of the order of 2 mV. It is interesting that a similar phenomenon was observed without stirring both in a two-phase system consisting of organic and aqueous solutions and in a liquid membrane system consisting of aqueous, organic and aqueous solutions, and that the amplitudes of the oscillations in the latter system were as much as two orders of magnitude more than that in the former system. It should be noted that in these experiments the concentrations of the solutes were far-from-equilibrium, i.e., the hydrophilic substance, picric acid, was dissolved in the organic phase and the hydrophobic substance, CTAB, was dissolved in the aqueous phase. Dupeyrat and Nakache [11,12] have observed quasi-periodic variations of the relaxation type in the interfacial tension under similar experimental conditions. In addition, the presence of an unstirred layer seems essential for generating electrical oscillations. It should be noticed that the presence of 'unstirred' layers has been considered to play an important role in generating 'excitation' in biological membranes [13].

Teorell [4] showed that a porous membrane with fixed charges under a hydrostatic pressure difference, through which a constant electric current was passed, could generate oscillation of electrical potential. Mueller and Rudin [6] found that when artificial lipid membranes were modified by some proteinaceous substances, they spontaneously exhibited rhythmic electrical oscillations. It has been reported by Kobatake [8] that a Millipore membrane containing a synthetic lipid analog, dioleyl phosphate, showed excitability, i.e., oscillation of electrical potential across the membrane. Pant and Rosenberg [7] reported that sustained oscillations were generated in bilayer lipid membranes in the presence of $K_3Fe(CN)_6$ and KI in the aqueous bathing solution [7]. In the present study, we showed that the oscillations of electrical potential occurred spontaneously in a 'thick' artificial liquid membrane. The amplitudes of the oscillations in our experiments were much greater than those reported by Mueller and Rudin ($\approx$ 40 mV [6]), Kobatake ($\approx$ 20 mV [8]) and Pant and Rosenberg ($\approx$ 20 mV [7]). Further experimental and theoretical studies on oscillatory phenomena in the liquid membrane system are in progress in our laboratory.

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