

Contact Phenomena at the Heptane|Water Interface in the Presence of Valinomycin *

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Summary

The influence of valinomycin on the change of the VOLTA potential in the water|heptane system has been studied by means of a vibrating capacitor and a radioactive probe. The results obtained are compared with the e.m.f. of an electrochemical concentration circuit simulating a membrane. The non-additivity of the potentials of the two half-cells heptane|water, observed when a transmembrane potential arises in the presence of valinomycin, is associated with the appearance in the heptane phase of a compensation potential.

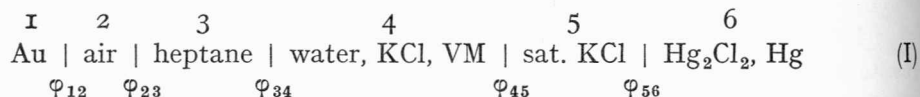
Two effects have been found to exist in the system investigated. The first effect (the unipolar charging of heptane in equilibrium with the water phase in the presence of valinomycin) increases with the lowering of the temperature and at temperatures close to 0 °C can reach 18 V. The second effect (additional electrization of the heptane layer containing the space charge) is recorded when a vibrating gold reference electrode is immersed into the nonaqueous phase and the amplitude of vibration exceeds a certain limit and seems to be associated with electrokinetic phenomena.

Introduction

The presence of valinomycin (VM) makes a bimolecular lipid membrane permselective with respect to K^+ .¹ A detailed study of the transmembrane potential in a concentration circuit with the use of specially prepared electrodes, based on VM and selective with respect to the K^+ , showed^{2,3} that the potential of the concentration circuit depends linearly on the logarithm of the K^+ activity with the slope 58 mV. The concentration circuit of the type "water|oil|water" contains two interfaces between aqueous and non-aqueous phases, or, as we shall call it in what follows, two half-cells of the type "water|oil". As a model of such a

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half-cell we chose the system heptane|water, on which we studied the change in the interfacial potential upon introduction of VM into the system.^{4,5} The technique used enabled to measure by means of a vibrating capacitor the change in the VOLTA potential (φ) between an Au reference electrode and heptane, caused by a change in the composition of the aqueous phase in the cell¹



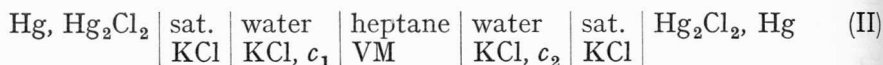
The set-up used consisted of a Pyrex cell with a bowl, in which heptane lay over the aqueous phase in accordance with the specific gravity of these liquids. Above the heptane phase the vibrating gold reference electrode was placed. In the aqueous phase, a calomel electrode was used as a reference electrode. It was connected with the cell by a salt bridge with saturated KCl solution. All aqueous solutions had been prepared from doubly distilled water and twice recrystallized salts. Heptane was of normal standard grade. It was placed in the same vessel as the aqueous phase, where it remained 36 hours, or less if the mixture had been previously shaken on a rocker. Valinomycin was introduced as a highly concentrated solution in heptane or alcohol. If, upon a change in the composition of the aqueous phase 4 the potential differences φ_{12} , φ_{23} , φ_{45} and φ_{56} remained unchanged, the total observed effect should be ascribed to the change in the potential difference φ_{14} at the heptane|water interface. It was shown by special experiments on the system



that the Volta potential remains unchanged with varying heptane phase composition. The heptane phase consisted of heptane separated from the previously equilibrated system heptane|aqueous solution. Actually, the constancy of the potential jump φ_{23} means that there occur no changes in the surface potential at the heptane|air interface. Moreover, the absence of adsorption was confirmed by measurements of the heptane surface tension at the interface with air. The constancy of φ_{12} means that the change in the VOLTA potential is not due to a change in the electron work function of Au caused by increase or decrease of heptane adsorption on its surface. Thus, the value of $\Delta\varphi$ reckoned from the supporting electrolyte potential, can be referred to the water|heptane interface which is being investigated.

The cell (I) was investigated also by means of a radioactive Pu probe, which ionized the space between the Au gauze and the aqueous phase.

The electrochemical concentration cell simulating the membrane contained heptane and VM:



The concentration c_1 remained constant ($10^{-2} M$ KCl), while the concentration c_2 varied, as shown in Fig. 1a. The e.m.f. of the cell (II) taken as a function of the KCl concentration in the presence of $10^{-6} M$ VM has the theoretical slope 58 mV for a tenfold increase of K^+ concentration. The cell (II) can be divided into 2 half-cells (cell I). The VOLTA potential of each half-cell was measured by means of a vibrating capacitor (VC). In Fig. 1b the Volta potential is plotted versus the K^+ concentration in the presence of VM as referred to the supporting electrolyte, KCl solution without VM. As it is clear from the comparison of Fig. 1a with 1b, the potentials of the two half-cells heptane|water are not additive. This non-additivity is evidently associated with the appearance of a potential drop within the heptane phase of cell (II).

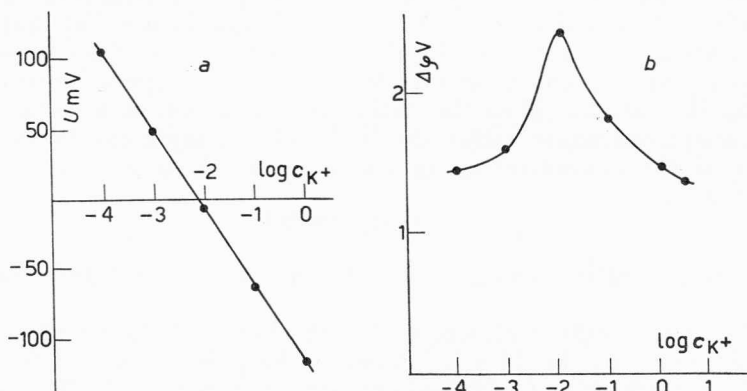
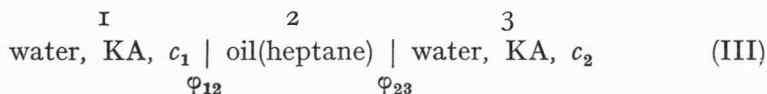


Fig. 1.

Dependence of transmembrane and interfacial potentials on the K^+ ions concentration in the presence of $10^{-6} M$ VM. *a*: cell (I); *b*: cell (II).

Let us consider the establishment of the potentials in the system



in the simplest case when the same salt KA is dissolved in the aqueous phases 1 and 3. If at the interfaces 1|2 and 2|3 equilibrium is established for cation and anion, it can be expressed in terms of the equality of the electrochemical potentials of individual ions in the aqueous $\tilde{\mu}^{\text{aq}}$ and nonaqueous $\tilde{\mu}^{\text{oil}}$ phases. If we supplement this by the condition of electroneutrality of each of the phase and consider only one interface between the semiinfinite phases oil|water, we can obtain the usual for-

mula relating the distribution coefficients oil-water of cation B_K and anion B_A to the potential difference at the oil-water interface

$$\varphi_r = \frac{RT}{2F} \ln \frac{B_K}{B_A}$$

where

$$\ln B_K = \frac{{}^o\mu_K^{\text{aq}} - {}^o\mu_K^{\text{oil}}}{RT}; \quad \ln B_A = \frac{{}^o\mu_A^{\text{aq}} - {}^o\mu_A^{\text{oil}}}{RT} \quad (1)$$

In what follows, in considering the total cells I-III we shall not make use of the electroneutrality condition in the form of the equality of the cations and anions concentrations in the nonaqueous phase, but will assume that when nonequilibrium (adsorption) potentials arise at the interfaces 1 | 2 and 2 | 3, there exists still equilibrium distribution for the penetrating ion (*i.e.* the ion to which a greater value of B corresponds) in a certain layer adjoining the interface. For concreteness, let us assume this to be the cation. Then the cation concentration at a certain point of the nonaqueous phase within the limits of this layer can be expressed in terms of the concentration in the aqueous phase at the interfaces 1 | 2 and 2 | 3.

$$c_K^{\text{oil}} = c_K^{\text{aq}} B_K e^{-nF\varphi/RT} \quad (2)$$

where n is the cation charge, φ the oil | water potential difference (φ_{23} or $-\varphi_{12}$).

When the circuit is closed, within the limits of the oil phase a potential difference φ_c should arise between the points near the interfaces 1 | 2 and 2 | 3, which can be called compensation potential. This ensures equilibrium within the oil phase for the penetrating ion. We shall not introduce an equivalent assumption for the anion.

The equilibrium condition in the oil phase can be written as

$$nF\varphi_c = {}^2\mu_K^{\text{oil}} - {}^1\mu_K^{\text{oil}}$$

$$\varphi_c = \frac{RT}{nF} \ln \frac{{}^2c_K^{\text{oil}}}{{}^1c_K^{\text{oil}}} \quad (3)$$

where μ_K^{oil} designates the chemical potential of the cation, the superscripts 2 and 1 referring to the interfaces 2 | 3 and 1 | 2, respectively. Using relation (2), we can substitute the cations concentration in the oil phase by those in the aqueous phase

$$\varphi_c = -\varphi_{12} - \varphi_{23} + \frac{RT}{nF} \ln \frac{{}^2c_K^{\text{aq}}}{{}^1c_K^{\text{aq}}} \quad (4)$$

The nature of the compensation potential depends on the oil phase. It can arise due to polarization in dielectric liquids, which contain colloidal aggregates including water molecule, or can be a diffusion potential for

well-conducting media. This problem requires further investigation in each particular case.

The total value of the potential difference between aqueous phases 1 and 3 at the ends of the concentration cell φ_s consists of three terms

$$\varphi_s = \varphi_{12} + \varphi_{23} + \varphi_c \quad (5)$$

and with account taken of (4)

$$\varphi_s = \frac{RT}{nF} \ln \frac{{}_2c_{K^{aq}}}{{}_1c_{K^{aq}}} \quad (6)$$

It is significant that the compensation current responsible for the appearance of φ_c is proportional to the ions concentration in the oil phase, and on account of their low concentration and the small capacity of the system, can be extremely small.

The physical sense of the conclusion given here, essentially a trivial one, lies in the elucidation of the fact that the setting up of the potential difference φ_s between the aqueous phases 1 and 3, which equilibrates the system with respect to the cation, does not necessitate the establishment of the equilibrium distribution potentials at the interfaces oil|water, if the electroneutrality condition is not fulfilled in the oil phase.

The same expression for φ_s can be obtained by assuming that equilibrium distribution potentials are set up at interfaces 1|2 and 2|3, but that the transference number of the cation t_K in the oil phase is unity.

To understand the reasons of the selectivity of cell (II) with respect to the K^+ ion, we studied in detail the VOLTA potentials of the half cells (cell 1) containing chlorides of various cations. Fig. 2 shows the values of for Li^+ , Na^+ , K^+ , Rb^+ , Cs^+ cations referred to the Na^+ potential.

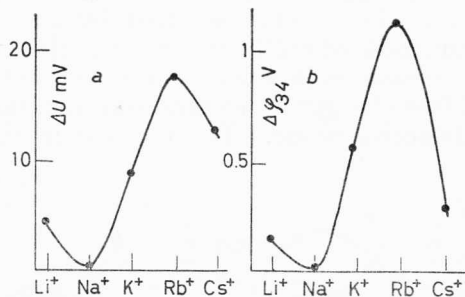


Fig. 2.

Dependence of transmembrane and interfacial potentials on the nature of cations at the MeCl concentration $10^{-4} M$ in the presence of VM. *a*: biionic potentials of bilayer phospholipid membranes, according to the data of ref. 6-8; *b*: interfacial potentials relative to the potential in the case of Na^+ .

The comparison of the biionic potentials of phospholipid membranes⁶⁻⁸ with the potentials measured at the same concentrations of alkali metals chlorides in the presence of 10^{-6} M VM shows that in both cases the shape of the curves of the potential difference versus the cation nature is similar, but in the case of the cells with one interface the absolute values are nearly by 2 orders of magnitude higher. The VOLTA potentials for the systems containing K^+ and Rb^+ ions have the highest values.

Addition of enanthic acid to the heptane phase at pH values of aqueous solution greater than pK of enanthic acid increases the VOLTA potential due to the presence of VM, whereas at $pH < pK$, it reduces the VOLTA potential down to the values characteristic of the system in the absence of VM (Fig. 3). The presence of an organic anion seems to increase the adsorption of the complex $K(VM)^+$, whereas strongly adsorbed undissociated enanthic acid molecules displace it from the interface.

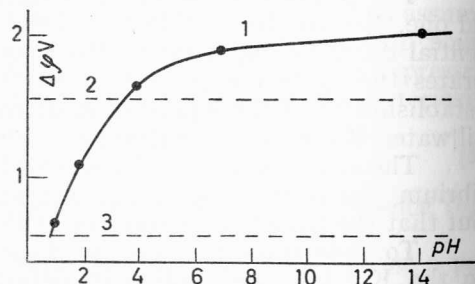


Fig. 3.

Influence of pH on the change in the interfacial potential in the system heptane|water in the presence of 1: 10^{-2} M KCl + 10^{-6} M VM + 10^{-3} M enanthic acid; 2: 10^{-2} M KCl + 10^{-6} M VM; 3: 10^{-2} M KCl.

We gain better insight into the nature of high potentials observed at the water|heptane interface if we compare the results of the Volta potential measurements of cell (I) by means of the vibrating capacitor and the radioactive probe methods. As it follows from Fig. 4, in both cases the dependences of the VOLTA potential on KCl concentration are non-monotonic. However, the effects measured by the radioactive probe method lie within hundreds of millivolts, whereas the vibrating capacitor method gives values measured in volts. This is accounted for by the fact that the number of free charges in heptane was artificially increased due to the use of a radioactive probe. The increase in the number of free

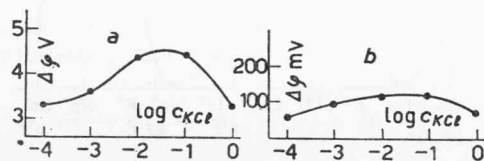


Fig. 4a,b.

Dependence of VOLTA potential on the K^+ concentration in cell (I) (10^{-7} M VM). a: cell (I), the vibrating capacitor method; b: cell (I), the radioactive probe method.

charges can be also responsible for the lesser effect observed with increased KCl concentrations. Apparently charged aggregates accumulate in heptane, which include ions $(KVM)^+$, water molecules and, possibly, some counterions. The nature of the particles determining the charging of heptane is not yet quite clear. The unipolar charging of the heptane phase occurs until the arising electric field begins to hinder further penetration of the charges into heptane.

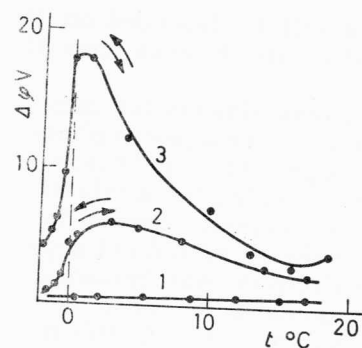


Fig. 5.

Dependence of VOLTA potential in cell (I) on temperature: 1: 10^{-2} M KCl; 2: 10^{-2} M KCl + 10^{-7} M VM; 3: 10^{-2} M KCl + 10^{-6} M VM.

The electrization effect detected by us was observed also during the measurement of the temperature dependence of the VOLTA potential in cell (I). Fig. 5, 1 shows the temperature-independence of the VOLTA potential in cell (I) in the case of a system without VM. However, already at a 10^{-7} M VM content (Fig. 5, 2) the quantity being measured de-

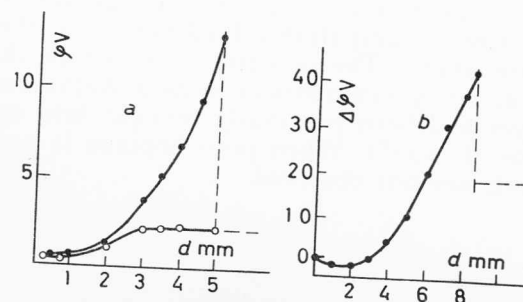
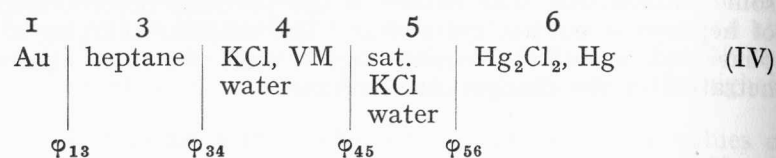


Fig. 6

Dependence of potential in the system heptane|water, measured by the vibrating capacitor method, on the heptane layer thickness at amplitudes 1 mm (1), 0.1 mm (2). a: 20°C , b: 0°C . Dashed line: cell (I), $d > 5$ mm, solid line: cell (IV).

depended essentially on temperature. At $10^{-6} M$ VM (Fig. 5, 3), the effect in cell (I) reaches 18 V at a temperature close to zero. If the vibrating electrode was directly immersed in heptane (cell IV)



the results of measurements, unlike those of cell I, depended on the amplitude of the Au electrode vibration and on its distance from the water surface, d .

Fig. 6 a shows the results of one of the experiments for cells (I) and (IV) at $10^{-2} M$ KCl and $10^{-6} M$ VM content in the aqueous phase 4, with which heptane had been previously brought into equilibrium, at the maximum (curve 1) and minimum (curve 2) amplitudes of vibrations of the reference electrode. When the reference electrode vibrated in air (cell I, $d > 5$ mm), the VOLTA potential value measured did not depend on the amplitude of vibrations. When the electrode was immersed in heptane (cell IV), the result of the measurement depended on the amplitude of vibrations until it became less than 0.1 mm. At this minimum amplitude the results of the potential measurements for cells (I) and (IV) with the Au electrode near the free surface of heptane are similar.

At the maximum amplitude of vibrations, the potential value measured increases with increasing d without any sign of saturation and in the above experiment reached 13 V near the heptane surface.

With decreasing temperature, the heptane electrization at large amplitudes reached the enormous value of 40 V (Fig. 6 b). In this case the difference between the potential values in cells (I) and (IV) means that $\varphi_{12} + \varphi_{23}$ is not equal to φ_{13} .

The additional electrization of the heptane phase containing VM observed in this study seems to be associated with electrokinetic phenomena, and for its complete manifestation not only the presence of VM is necessary, but also an amplitude of vibrations of the Au electrode exceeding a certain limit. The potential recorded in this case can be observed in a completely symmetrical system Au|heptane|Au as well, if the heptane layer had been previously brought into equilibrium with the aqueous phase of cell (I). When pure heptane is used in the same system, these effects are not observed.

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