

Pore formation by alamethicin in planar bilayers below lipid phase transition temperature

W. Hanke\*, H. Eibl\*\*, and G. Boheim\*

\*Abt. Zellphysiologie, Ruhr-Univ. Bochum, D-4630 Bochum

\*\*MPI Biophysikalische Chemie, D-3400 Göttingen

Planar bilayer membranes were formed according to the method of Montal and Mueller using 1-stearoyl-3-myristoyl-glycero-2-phosphocholine (1,3-SMPC). Lipid phase transition temperature is  $T_C = 29^\circ\text{C}$  for the cooling and  $T_C = 31^\circ\text{C}$  for the heating process. The properties of the 1,3-SMPC/alamethicin multi-pore and single-pore systems were studied within the temperature range  $14-40^\circ\text{C}$ .

At constant applied voltage the current/temperature characteristic of the multi-pore system shows a pronounced maximum at  $\sim 24^\circ\text{C}$  and a shoulder at  $\sim 18^\circ\text{C}$ . These two effects are explained in terms of single-pore parameters in the following way.

1. the maximum: A second population of alamethicin pores with anomalously long mean pore lifetime ( $\tau_p^* \geq 10\text{s}$ ) is observed between  $24-30^\circ\text{C}$  besides the usually observed population of short living pores ( $\tau_p \sim 0.4\text{s}$ ). The maximum current induced by this second population is found at  $\sim 27^\circ\text{C}$ . The simultaneous existence of two different pore populations at  $T \approx T_C$  seems to result from a lateral separation of the membrane components into a fluid 1,3-SMPC/alamethicin phase and into a frozen 1,3-SMPC phase containing only a small amount of alamethicin.

2. the shoulder: Pore state conductances  $A_p$  show a temperature dependence according to an activation energy of  $\sim 25\text{ kJ mol}^{-1}$  within the considered temperature range. The mean pore lifetime  $\tau_p$ , on the other hand, increases with decreasing temperature. This increase is stronger below  $21^\circ\text{C}$  than above. The pore formation rate  $f_p$  of the short-living population shows maximum values in the range  $27-22^\circ\text{C}$  and decreases steeply below  $\sim 19^\circ\text{C}$ .

Single-pore and multi-pore data are consistent, if one takes into consideration the physico-chemical rule that a larger concentration of the solved component (alamethicin) leads to a larger freezing point depression of the solution.