The Stabilizing Effect of Ion Occupancy on the Formation of Membrane Channels*

JOHN SANDBLOM, AVI RING and EDGAR DIAZ**

Inst. of Physiol. and Med. Biophysics, Biomedical Centre, P.O. Box 572, S-751 23 Uppsala, Sweden

A well-characterized channel is that formed by the neutral polypeptide, gramicidin A, which dimerizes in lipid bilayers to form channels with a diameter of 0.4 nm.[1] Studies on the binding of ^{23}Na to malonyl gramicidin in lysolecithin micelles have shown the presence of at least two binding sites[2] whereas the results of electrical measurements[3,4] in combination with flux-ratio determinations[5] suggest the extension to four binding sites.[6] By fitting a 4-site model with low central and low outer barriers (*i.e.* the two middle barriers are rate limiting) to conductance data for CsCl, the successive binding constants for loading the channel with one to four cesium ions are calculated to be respectively 500 M^{-1}, 2 300 M^{-2}, 60 M^{-3} and 900 M^{-4}.

Since the channel is symmetrical, the even numbered occupancies (2 and 4) represent symmetrical configurations of the channels. These configurations are seen to be energetically favoured over the asymmetrical configurations. This suggests that the loading of ions are associated with configurational changes of the monomers and it is further suggested that symmetrical loading of ions should have a stabilizing effect on the channel formation.

We have tested this prediction by comparing the symmetrical occupancies with the life times of the channel. Fig. 1 shows the probability curve calculated for symmetrical occupancies and the points are measured life times[7] scaled to coincide with the theoretical curve at high concentrations. The increase in channel life times is then seen to follow the isotherm of symmetrically loading the channel.

1. Urry, D. N. *Proc. Natl. Acad. Sci. U.S.A. 68* (1971) 672.
2. Urry, D. N., Venkatachalam, C. M., Spisni, A., Bradley, R. J., Trapane, T. L. and Prasad, K. U. *J. Membr. Biol. 55* (1980) 29.
3. Sandblom, J., Eisenman, G. and Neher, E. *J. Membr. Biol. 31* (1977) 383.
4. Neher, E., Sandblom, J. and Eisenman, G. *J. Membr. Biol. 40* (1978) 97.
5. Procopio, P. and Andersen, O. S. *Biophys. J. 25* (1979) 8.
6. Sandblom, J., Eisenman, G. and Hägglund, J. *To be published.*
7. Kolb, H. A. and Bamberg, E. *Biochim. Biophys. Acta, 464* (1977) 127.

Received November 20, 1980.

* Communication at the Joint Meeting of the Swedish Biochemical and Swedish Biophysical Societies in Uppsala, 28–29th November, 1980.

** Permanent address: Department of Physics, Universidad del Valle, Cali, Colombia.

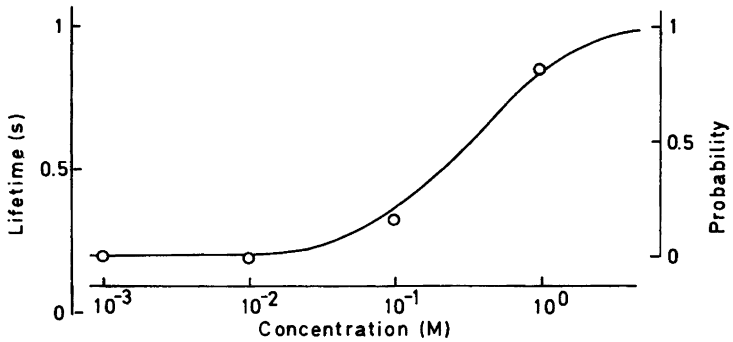

Fig. 1. The probability that the channel is occupied by two or four cesium ions is plotted against the concentration of CsCl in the bathing solution. The curve was calculated from binding constants and the points represent direct measurements of the channel life times.[7]

0302-4369/81/010060-01$02.50