Dimer versus tetramer

Dear Sir:

We see no convincing evidence in favor of an aggregational model for the gramicidin channel, and find the evidence in favor of a (generalized) dimer model to be overwhelming. Many of the arguments were summarized or discussed by Cifu et al. (1992); here we wish to make three points.

(*a*) From the distribution of single-channel current transition amplitudes it can be concluded that gramicidin channels have a single predominant conformation, i.e., being either dimers or tetramers (or another oligomer of defined molecularity) but not some less defined aggregate (cf. Andersen et al., 1987; Sawyer et al., 1989; Cifu et al., 1992).

(*b*) In hybrid channel experiments, one sees at most two hybrid channel types (Apell et al., 1977; Veatch and Stryer, 1977; Andersen et al., 1988; Cifu et al., 1992), and the hybrid channel appearance rates and energetics are consistent with the formation of dimers but not with the formation of tetramers (Veatch and Stryer, 1977; Durkin et al., 1990). These results can only, with serious difficulties, be reconciled with any type of lateral aggregation model.

(*c*) Both the relaxation kinetics and the sequence-dependent changes in channel-forming potency can be accounted for by a generalized dimer model (Becker et al., 1991; Cifu et al., 1992). In contrast, it is not clear how to reconcile the very different single-channel durations of [Val^1]gramicidin A and malonyl-bis-gramicidin channels (cf. Bamberg and Janko, 1977) with similar relaxation behaviors reported by Stark et al. (1986). That the malonyl-bis-gramicidin sample used by Stark et al. gave rise to several classes of channel events suggests contamination.

REFERENCES

Andersen, O. S., J. T. Durkin, and R. E. Koeppe II. 1988. Do amino acid substitutions alter the structure of gramicidin channels? Chemistry at the single molecule level. *Jerusalem Symp. Quantum Chem. Biochem.* 21:115–132.

Andersen, O. S., R. E. Koeppe II, J. T. Durkin, and J.-L. Mazet. 1987. Structure-function studies on linear gramicidins: site-specific modifications in a membrane channel. *In* Ion Transport Through Membranes. K. Yagi and B. Pullman, editors. Academic Press, Tokyo. 295–314.

Apell, H.-J., E. Bamberg, H. Alpes, and P. Läuger. 1977. Formation of ion channels by a negatively charged analog of gramicidin. *A. J. Membr. Biol.* 31:171–188.

Bamberg, E., and K. Janko. 1977. The action of a carbon suboxide dimerized gramicidin A on lipid bilayer membranes. *Biochim. Biophys. Acta.* 465:486–499.

Becker, M. D., D. V. Greathouse, R. E. Koeppe II, and O. S. Andersen. 1991. Amino acid sequence modulation of gramicidin channel function. Effects of tryptophan-to-phenylalanine substitutions on the single-channel conductance and duration. *Biochemistry.* 30:8830–8839.

Cifu, A. S., R. E. Koeppe II, and O. S. Andersen. 1992. On the supramolecular structure of gramicidin channels. The elementary conducting unit is a dimer. *Biophys. J.* 61:189–203.

Durkin, J. T., R. E. Koeppe II, and O. S. Andersen. 1990. Energetics of gramicidin hybrid channel formation as a test for structural equivalence. Side-chain substitutions in the native sequence. *J. Mol. Biol.* 211:221–234.

Sawyer, D. B., R. E. Koeppe II, and O. S. Andersen. 1989. Induction of conductance heterogeneity in gramicidin channels. *Biochemistry.* 28:6571–6583.

Stark, G., M. Strässle, and Z. Takacz. 1986. Temperature-jump and voltage-jump experiments at planar lipid membranes supported an aggregational (micellar) model of the gramicidin A ion channel. *J. Membr. Biol.* 89:23–37.

Veatch, W., and L. Stryer. 1977. The dimeric nature of the gramicidin A transmembrane channel: conductance and fluorescence energy transfer studies of hybrid channels. *J. Mol. Biol.* 113:89–102.

Olaf S. Andersen* and Roger E. Koeppe II[§]

**Department of Physiology and Biophysics*
Cornell University Medical College
New York, New York 10021 USA
and
[§]Department of Chemistry and Biochemistry
University of Arkansas
Fayetteville, Arkansas 72701 USA

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