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Supporting Material

Understanding ion conductance on a molecular level: An all-atom modeling of the bacterial porin OmpF

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Supplementary material for the manuscript Understanding ion conductance on a molecular level: An all-atom modeling of the bacterial porin OmpF

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I. DETAILS OF EXPERIMENTAL SETUP

The values of the conductance are determined with histograms on which a Gaussian distribution is fitted. The reported current is given by the peak position the corresponding Gaussian distribution. Furthermore, the conductance values reported here were determined only for the fully open state of the channel and not for transients which correspond to (partial) gating of the channel. Since OmpF is a trimer gating leads to a closure of individual monomer channels and therefore by a reduction of the current by 1/3, 2/3 or a complete closing of the channel. For the case of 83.5 ℃ the open state has a conductance of about 1200 pA. The transient states corresponding to closed channels then have currents of around 800 pA, 400 pA and 0 pA. Gating is observed for OmpF also at room temperature but at higher voltages.

For the selectivity measurements, aqueous solutions of KCl were buffered by 20 mM MES at pH 6. The membrane potential was applied using Ag/AgCl electrodes in 2 M KCl, 1.5% agarose bridges assembled within standard 200 μl pipette tips. Potential is defined as positive when it is greater at the side of protein addition (the cis side of chamber). The reversal potential was obtained as follows. In a first step, a lipid membrane was formed at a concentration gradient: 1 M KCl cis-side, 0.1 M KCl trans-side. In a second step, a single OmpF channel was inserted at zero potential and the channel conductance was checked by applying 50 mV and then potential polarity was switched. Finally, the ionic current through the channel was manually set to zero by adjusting the applied potential and thus obtaining the reversal potential. Each point was measured for at least three different OmpF samples to assure reproducibility and to estimate the standard deviation. Special care was taken to exclude any exchange of electrolyte solutions between the cis and trans compartments before the insertion of a single channel. In each measurement a new membrane was formed in fresh salt solutions.

Recently it was reported that the reversal potentials depend not only on the bathing solution content but also on the salt concentration in the agarose bridges [1]. For this reason Alcaraz et al. [1] assume that in the case of an asymmetric system the liquid junction potential should be accounted for. However under the current conditions this potential has a value of few mV only and is within the experimental error. In the present measurements the OmpF channel was inserted in a neutral lipid bilayer (DPhPC).

In Fig. S1 measured ion current traces for three different temperatures in an experiment with 100 mV applied voltage are shown. For example, at 83.5 ℃ the open state has a conductance of about 1200 pA. The transient states corresponding to one or two closed channels have currents of around 800 pA, 400 pA and 0 pA. In this context we should mention that gating of Ompf is also observed at room temperature but at higher voltages. However to date the underlying mechanism is unclear and our modeling does not reach typical time scales (second) in which gating occurs.

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[1] Alcaraz, A., E. M. Nestorovich, M. L. Lopez, E. Garcia-Gimenez, S. M. Bezrukov, and V. M. Aguilella, 2009. Diffusion, Exclusion, and Specific Binding in a Large Channel: A Study of OmpF Selectivity Inversion. *Biophys. J.* 96:56–66.

II. SUPPLEMENTARY TABLE OF CALCULATED CONDUCTANCES

T [℃]	[KCl] [M]	t_S [ns]	N_c	G [nS]	$\|\Delta G\|$ [nS]	$\|\Delta G/G\|$ [%]
2	0.5	10.0	67	1.06	0.13	13
12	0.5	10.6	100	1.51	0.15	10
22	0.5	10.0	87	1.39	0.15	11
37	0.5	10.0	114	1.82	0.17	10
57	0.5	10.7	156	2.34	0.19	9
72	0.5	10.5	199	3.03	0.22	8
80	0.5	10.0	208	3.32	0.23	7
90	0.5	10.0	229	3.56	0.24	13
2	1.0	12.9	118	1.46	0.14	10
12	1.0	10.0	96	1.54	0.16	11
22	1.0	12.2	189	2.49	0.18	8
37	1.0	12.4	311	4.02	0.23	6
47	1.0	10.8	313	4.63	0.27	6
57	1.0	10.4	313	4.80	0.28	6
72	1.0	10.6	386	5.9	0.3	6
80	1.0	10.0	340	5.5	0.3	6
90	1.0	10.0	433	6.9	0.4	5

TABLE S I: Details and results of the simulations including temperature T, simulation time t_S, crossing events N_c, conductance G and the error of the conductance $|\Delta G| = |G/\sqrt{N_c}|$.

III. SUPPLEMENTARY FIGURES

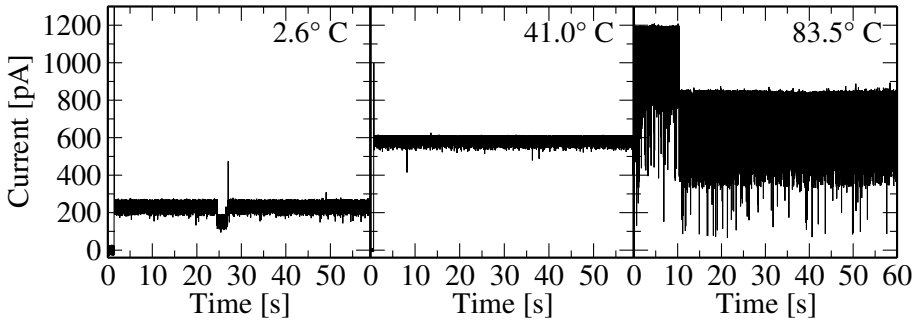

FIGURE S 1: Current through OmpF trimer in experiment with 100 mV applied voltage for 2.6, 41.0 and 83.5 ℃.

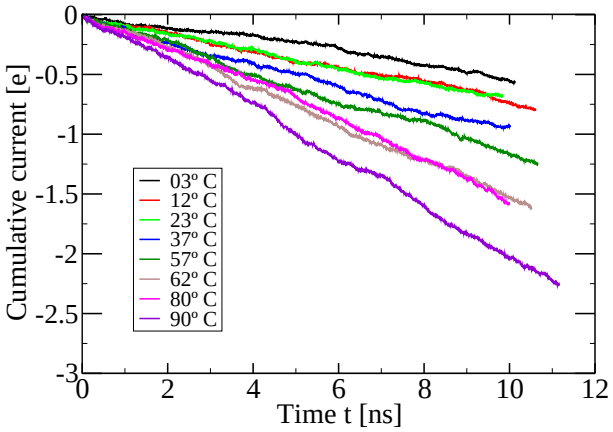

FIGURE S 2: Cumulative current through OmpF trimer. From the slope of the cumulative current the current is deduced.