Computer Simulation of a Rectifying Ion Channel

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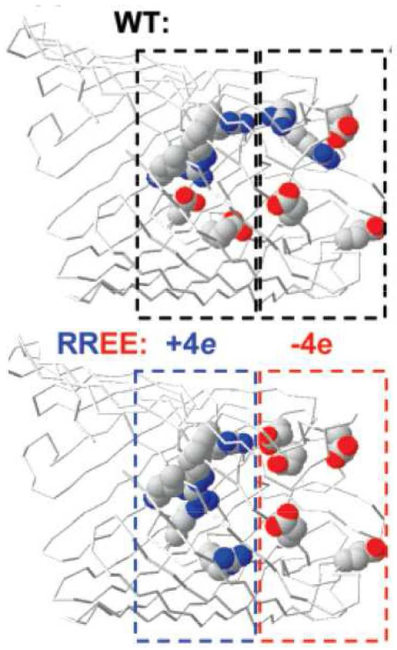
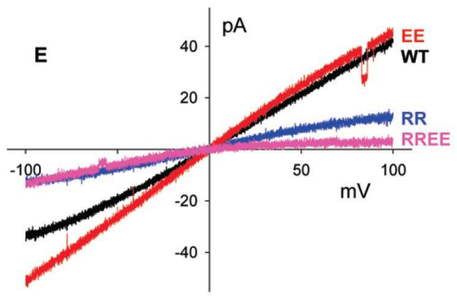
Aim

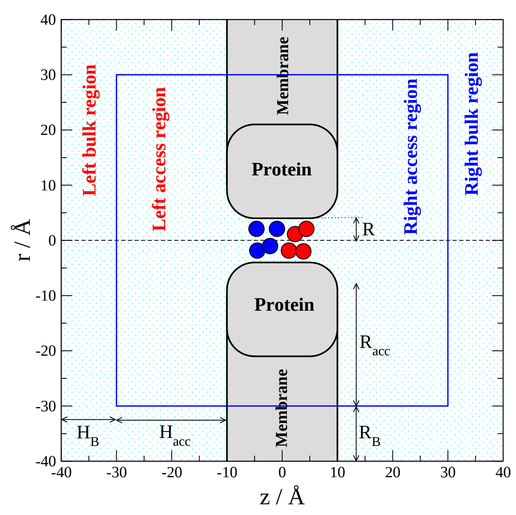
The goal of this work is to build models for the rectifying ion channel and study them with computer simulation methods thus trying to reproduce the phenomenon and to explain the mechanism behind it. The OmpF porin ion channel is a non-selective, non-rectifying in its wild-type (WT) form (top figure, source: [1]). Miedema et al. altered the distribution of amino acids to get a rectifying ion channel (figure in the middle). This mutated RREE channel has a charge distribution resembling a PN diode, so it was expected that it shows rectification behavior.

The behavior was found with electrophysiological measurements that provide the current through the channel at a given voltage (see RREE curve to bottom right).





Results with reduced model

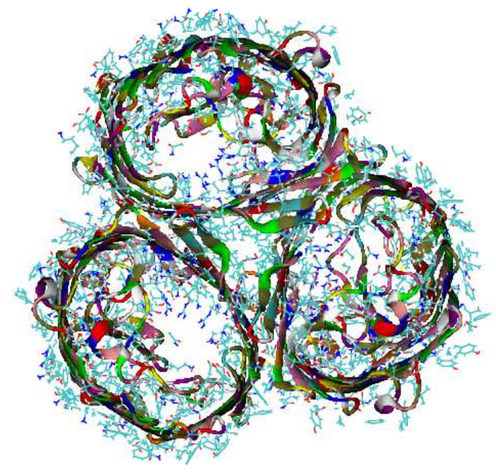


In the reduced model, we replaced the "important" charged residues with charged ions that are confined to certain region of the pore, but free to move inside. The rest of the channel protein is coarse-grained into a pore penetrating a membrane. Ions are modeled as charged hard spheres, the solvent is a dielectric continuum. Boundary conditions (concentrations and electrical potential) are set in the bulk regions on the boundary of the domain of solution. The access regions just outside the entrances of the channel are part of this domain.

The Nernst-Planck equation is solved for this domain. This equation provides the flux density if the concentration profiles and the electrochemical potential profiles are known. They are calculated with the Local Equilibrium Monte Carlo method [2,3].

Results with all-atom MD

- All-atom MD with the GROMACS[4] program suite
- External electric field of ±200 mV using periodic boundary conditions
- System size: ~11 nm × ~11 nm × ~11 nm → OmpF Porin trimer + + DMPC lipid bilayer + ions + water molecules: ~130 000 atoms
- Leap frog integrator (time step: 2 fs)
- Temperature coupling: Nose-Hoover
- Pressure coupling: Parrinello-Rahman
- Flexible models with position restraints for the protein backbone
- OmpF trimer structure: ProteinDataBank database (identifier: 2OMF)
- Protein/membrane complex generation: CHARMM-GUI [5]
- Sidechain mutation: VMD Mutate Residue plugin [6]



Snapshot of the OmpF trimer used in the MD simulations.

Simulation procedure:

Preliminary equilibration runs (no external electric field)

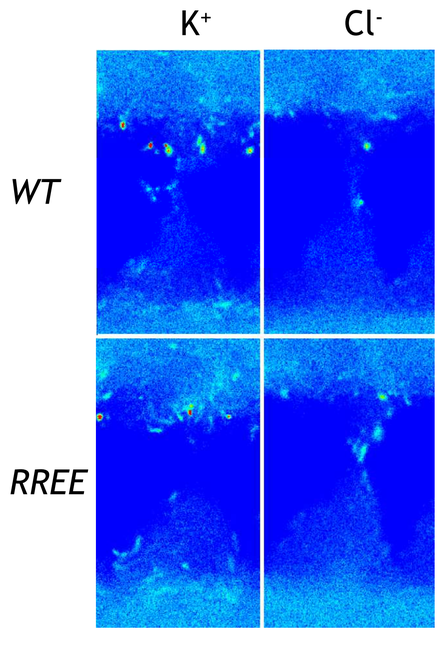
1.a. simulation box construction (preliminary energy minimization run)
1.b. 100 ps *NVT* run, T = 100 K,
1.c. 100 ps *NVT* run, T = 296 K
1.d. 1 ns *NpT* run, T = 296 K, p = 1 bar, isotropic pressure coupling
1.e. 3 ns *NpT* run, T = 296 K, p = 1 bar, semi-isotropic pressure coupling (independent coupling in the direction of transfer)

Simulations with applied external electric field at T = 296 K

2.a 10 ns *NVT* equilibration run
2.b 20 ns *NVT* production run

	Wild Type	RREE mutant
	I/pA	*I*/pA
+200 mV	216	152
-200 mV	-200	-176

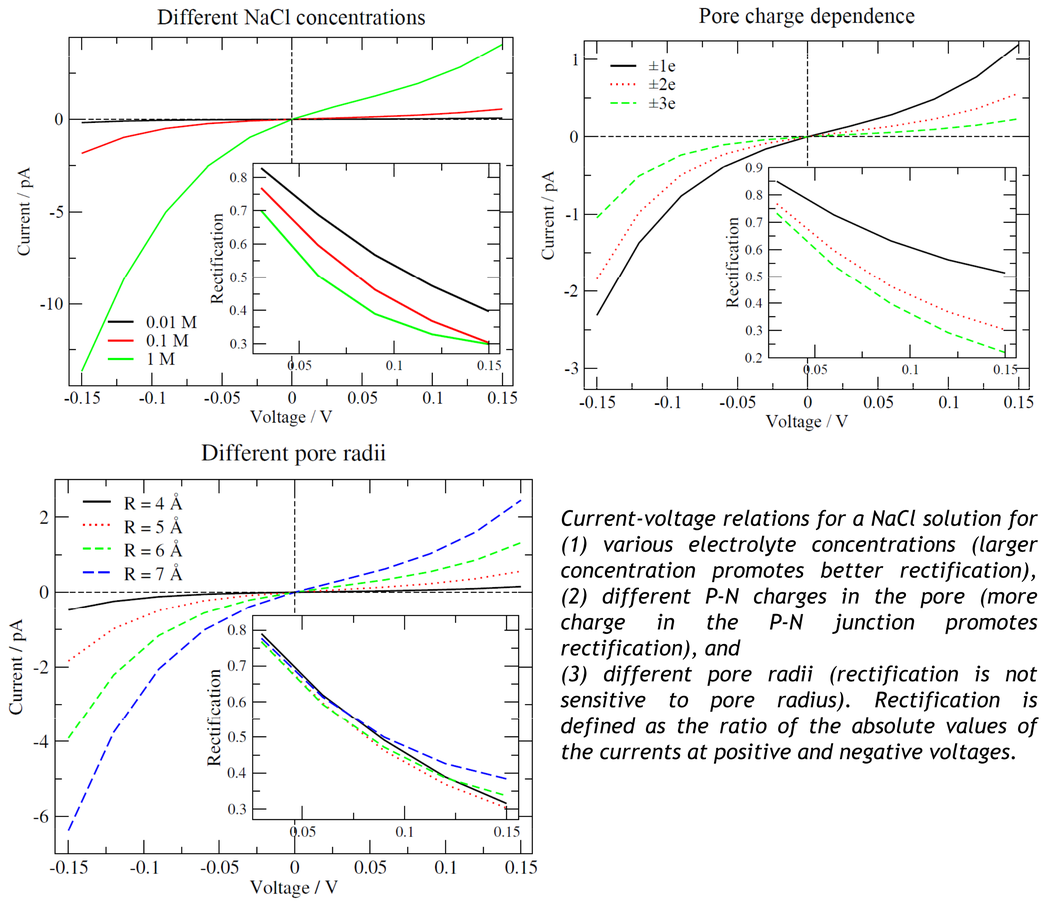
WT in 1 M KCl solution: qualitative agreement with experimental observations and literature [7]. *Simulations for RREE mutant: selectivity changes but no significant rectification.*



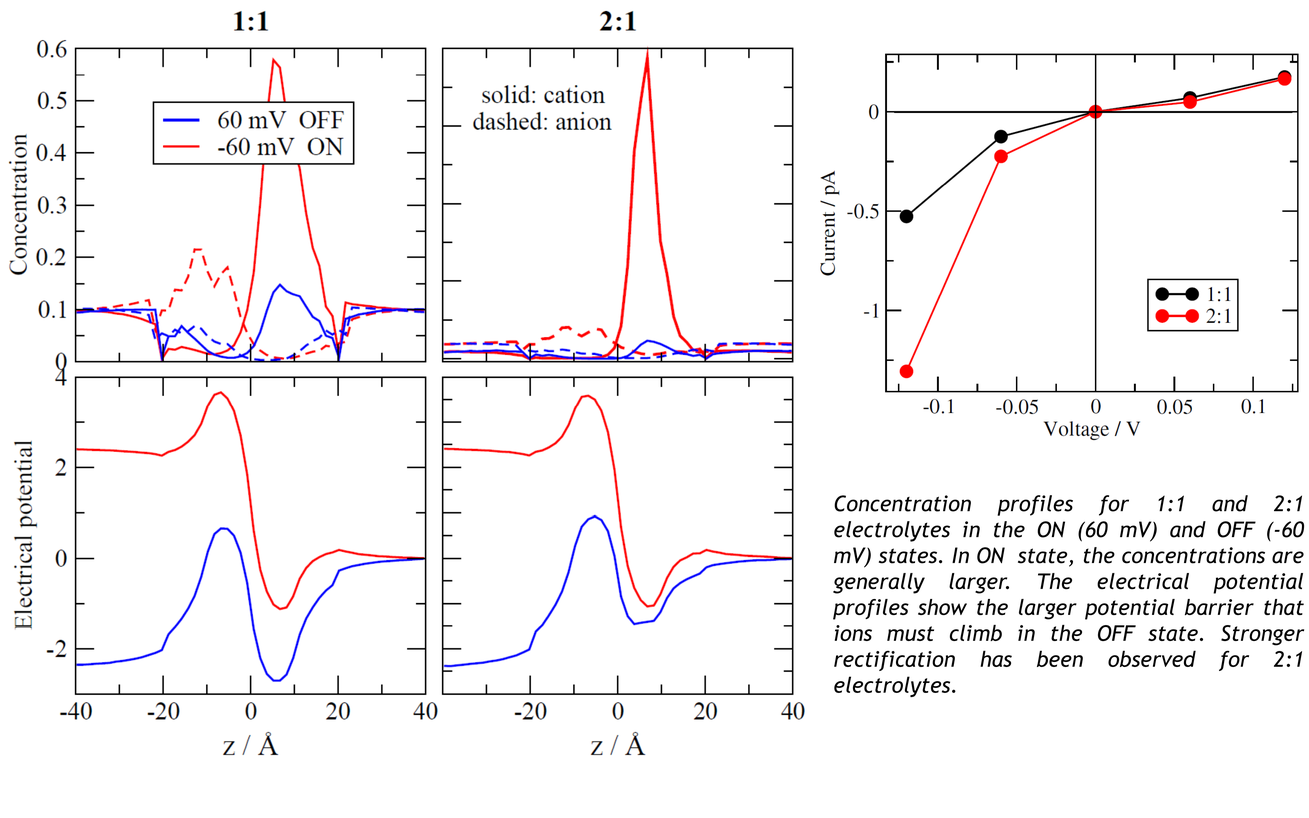
Ion density maps (accumulated for 20 ns) for single channel, obtained from simulation. Selectivity can be observed.

	Wild Type			RREE mutant		
	K+	Cl-	Σ	K+	Cl-	Σ
+200 mV	17	10	27	2	17	19
-200 mV	11	14	25	0	22	22

Total number of ions crossed the channel during the 20 ns production run.



Current-voltage relations for a NaCl solution for (1) various electrolyte concentrations (larger concentration promotes better rectification), (2) different P-N charges in the pore (more charge in the P-N junction promotes rectification), and (3) different pore radii (rectification is not sensitive to pore radius). Rectification is defined as the ratio of the absolute values of the currents at positive and negative voltages.



Concentration profiles for 1:1 and 2:1 electrolytes in the ON (60 mV) and OFF (-60 mV) states. In ON state, the concentrations are generally larger. The electrical potential profiles show the larger potential barrier that ions must climb in the OFF state. Stronger rectification has been observed for 2:1 electrolytes.

References

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Conclusion

The possibility of rectification in channels and nanopores is an intriguing possibility that opens new perspectives to bio- and nano-engineering. A mutated OmpF porin shows this behavior. Simulation studies are expected to yield insight to understanding this behavior. Interestingly, MD simulations for an all-atom model of the porin based on X-ray data does not reproduce the rectification behavior, while reduced models straightforwardly provide the results expected for a PN junction. The failure of the all-atom model motivates further examination. We are planning a multi-scale modelling project to fill the gap between these two limits of modelling level: we intend to build more structural detail into our reduced model.