Theoretical predictions of gating behavior for mutants of *Shaker*-type K_V channels from inter-domain energetics

Alexander Peyser† and Wolfgang Nonner‡

†Simulation Lab Neuroscience – Bernstein Facility Simulation and Database Technology,
Institute for Advanced Simulation, Jülich Aachen Research Alliance,
Forschungszentrum Jülich 52425 Jülich | Germany

‡Department of Physiology and Biophysics,
Miller School of Medicine, University of Miami,
Miami, Florida 33101 | USA

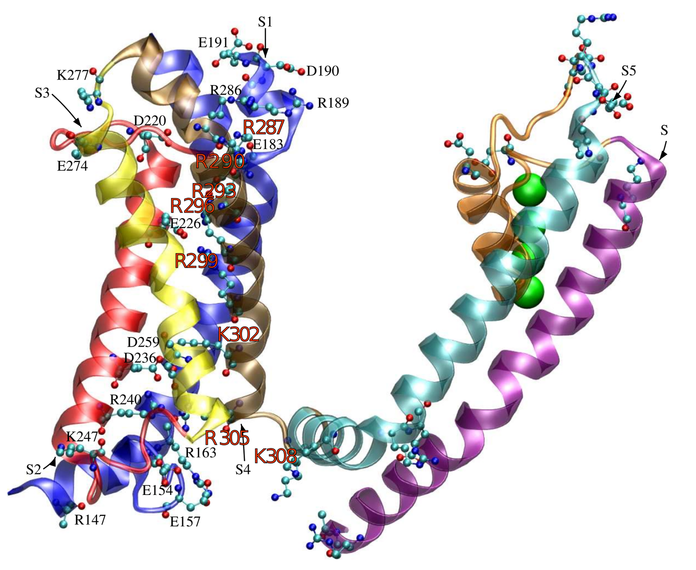
Abstract

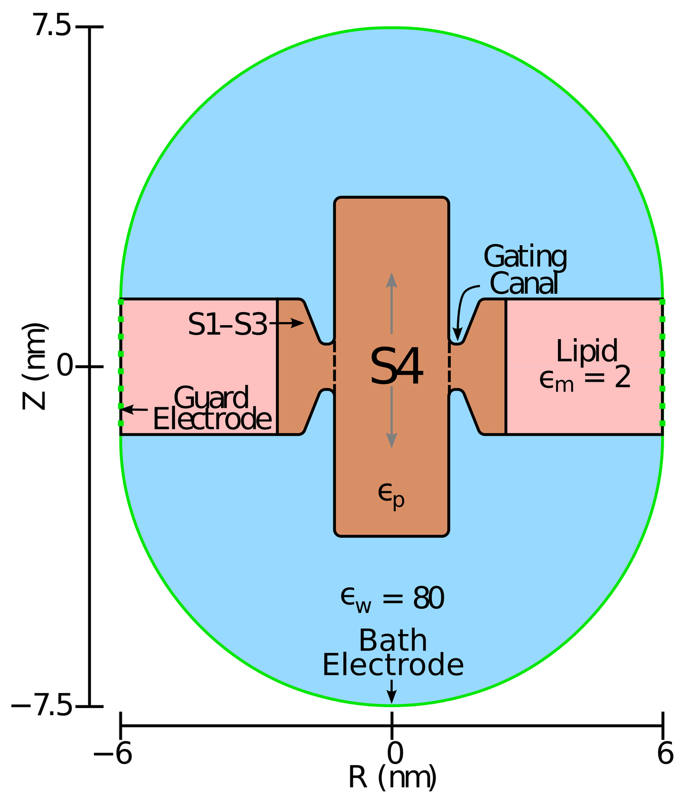
A multiscale physical model of *Shaker*-type K_V channels is used to span from atomic-scale interactions to macroscopic experimental measures such as charge/voltage (QV) and conductance/voltage (GV) relations. The model [1] comprises the experimentally well-characterized voltage sensor (VS) domains described by four replications of an independent continuum electrostatic model under voltage clamp conditions [2, 3] and a hydrophobic gate controlling the flow of ions by a vapor lock mechanism [4], connected by a simple coupling principle derived from known experimental results and trial-and-error. The total Hamiltonian of the system is calculated from the computed configurational energy for each components as a function of applied voltage, VS positions and gate radius, allowing us to produce statistical-mechanical expectation values for macroscopic laboratory observables over the full range of physiological membrane potentials ($|V| \leq 100$ mV, in 1 mV steps).

The *Shaker* QV and GV relations seen in Seoh et al. [5] are predicted by this model. With this approach, functional energetic relations can be decomposed in terms of physical components, and thus the effects of modifications in those elements can be quantified. We find that the total work required to operate the gate is an order of magnitude larger than the work available to the VS, and that the the experimentally observed bistable gating is due to the VS slide-and-interlock behavior.

The same model was systematically applied to VS charge mutants [5]. The QV and GV relations can be qualitatively predicted and the associated effects on functional domains determined. Additional features such as surface charges become significant for the pathological cases. Our engineering approach clearly elucidates that both normal function and mutant changes are electrostatic in nature.

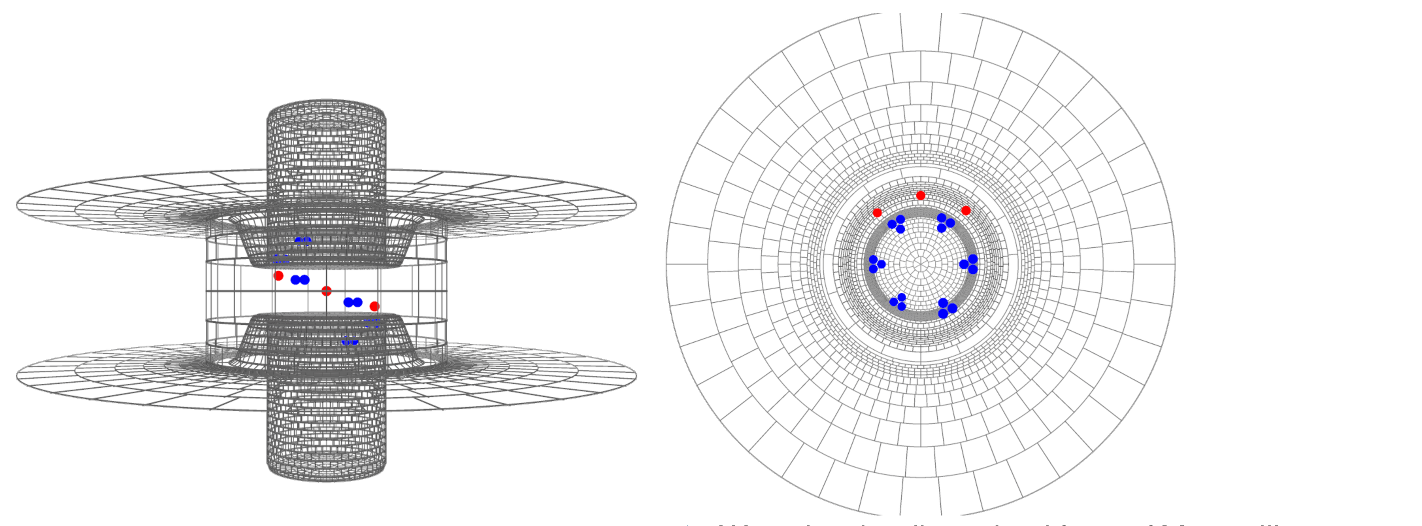
Voltage sensor of K^+ channels





(Above) Kv2.1 and Kv1.2 Chimera [6]: Kv are tetramers of this structure, S1–S4 is the voltage sensor, S5 & S6 for the conducting pore. Alignment of Chimera with *Shaker* of important residues [5] are R290:R362, R293:R365, R296:R368, R299:R371, K302:K374. (Right) Simulation cell of a single voltage sensor.

Discretized three-dimensional simulation cell of a single voltage sensor



$$4\pi\epsilon_0\,\mathbf{E}(\mathbf{r}) = \sum_k q_k^{\text{eff}} \frac{\mathbf{r}-\mathbf{r}_k}{|\mathbf{r}-\mathbf{r}_k|^3} + \int_{\mathcal{B}} \sigma^{\text{ind}}(\mathbf{r}') \frac{\mathbf{r}-\mathbf{r}'}{|\mathbf{r}-\mathbf{r}'|^3}\, da' + \int_{\mathcal{E}} \sigma^{\text{eff}}(\mathbf{r}') \frac{\mathbf{r}-\mathbf{r}'}{|\mathbf{r}-\mathbf{r}'|^3}\, da',$$

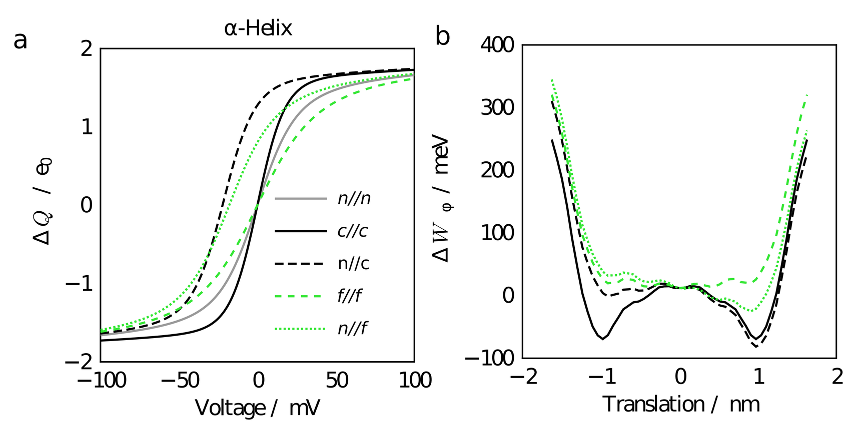
$$4\pi\epsilon_0\,V(\mathbf{r}) = \sum_k q_k^{\text{eff}} \frac{1}{|\mathbf{r}-\mathbf{r}_k|} + \int_{\mathcal{B}} \sigma^{\text{ind}}(\mathbf{r}') \frac{1}{|\mathbf{r}-\mathbf{r}'|}\, da' + \int_{\mathcal{E}} \sigma^{\text{eff}}(\mathbf{r}') \frac{1}{|\mathbf{r}-\mathbf{r}'|}\, da',$$

$$\sigma^{\text{ind}}(\mathbf{r}) = -\frac{\Delta\epsilon(\mathbf{r})}{\bar{\epsilon}(\mathbf{r})}\,\epsilon_0\,\mathbf{n}(\mathbf{r})\cdot\mathbf{E}(\mathbf{r}),$$

We solve the discretized form of Maxwell's equation using a surface element method (ICC):
$\mathbf{A}x = b$

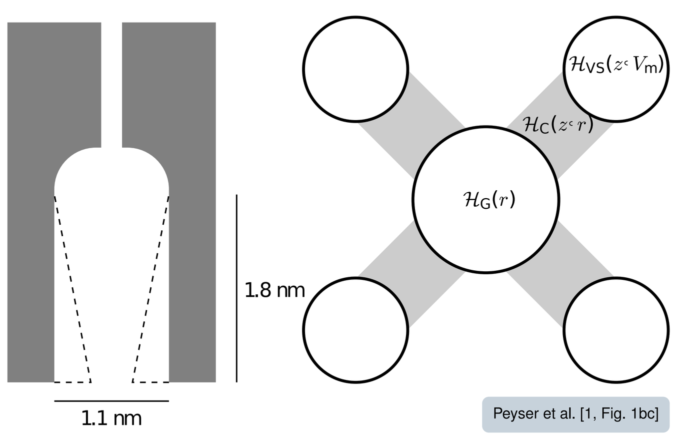
$\mathbf{A}$ are the interaction coefficients for $\mathbf{E}$ and V, x are the charges on tiles and fixed points, and b are the fixed potentials and mobile charges.

Consequences of surface charge for a single voltage sensor



Single VS model with discrete surface charge(s) added in four variations of geometrical position. The labels in panel a specify: n, no surface charge; c, charge in 'close' position; f, charge in 'far' position. (a) Mean charge–voltage relation; (b) Translational energy profile

Full system of 4 voltage sensors coupled to a hydrophobically-gated conducting pore

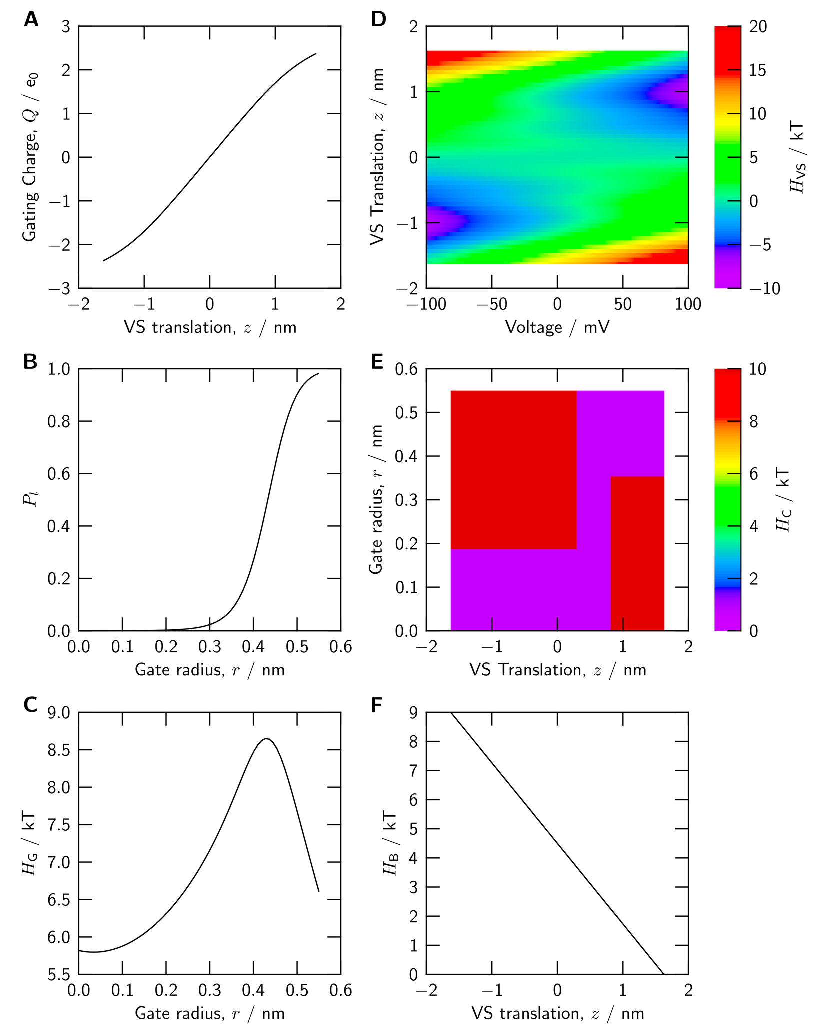


We combine 4 Hamiltonians to calculate the Boltzmann factors for every combination of VS position and gate radius.

- $\mathcal{H}_{\text{G}}(r)$ Potential energy of the gate with radius r [4]
- $\mathcal{H}_{\text{C},i}(z_i, r)$ Coupling function between one VS and the pore [1]
- $\mathcal{H}_{\text{B},i}(z_i)$ Work against an external force (preliminary representation of surface charge) [1]
- $\mathcal{H}_{\text{VS},i}(z_i, V_{\text{m}})$ Electrostatic potential energy of a z position for one VS at transmembrane potential V_{m} [2, 3]

$$B(\mathbf{z}, r, V_{\text{m}}) = \exp\left\{-\beta\left[\mathcal{H}_{\text{G}}(r) + \sum_{i=1}^{4}\left(\mathcal{H}_{\text{C},i}(z_i, r) + \mathcal{H}_{\text{B},i}(z_i) + \mathcal{H}_{\text{VS},i}(z_i, V_{\text{m}})\right)\right]\right\}$$

Charge displacement & Hamiltonians



Description of figures: [1, Fig. 2]

- A Charge displacement to physical translation mapping for wild-type VS
- B Gate radius to open probability mapping for morphometric model of hydrophobic gating. We define the conductance $G(r) = P_{\text{l}}(r)$, unless $r < 0.2$ nm where $G(r) = 0$
- C The Hamiltonian for the conducting pore
- D The Hamiltonian for the VS at a given translation and transmembrane potential for the wild-type VS
- E The hypothetical coupling Hamiltonian mapping VS positions and conducting pore radii to potential energies
- F A bias Hamiltonian, representing a simplified external interaction

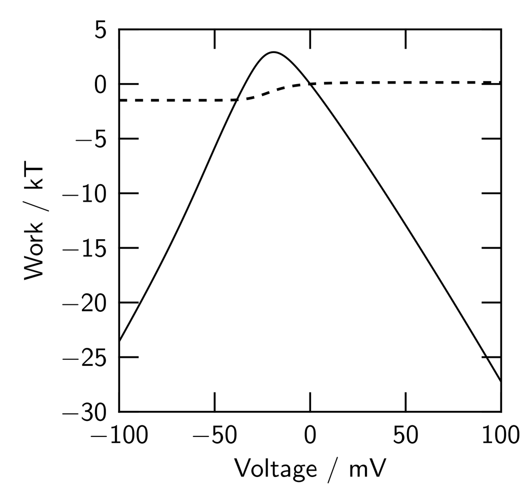
For Fig. C, the Grand Canonical energy is [4]
$\Omega_O = -p_l V + \sigma_l A + \kappa_l C$
$\Omega_c = -p_l V^l + \sigma_l A^l + \kappa_l C^l - p_g V^g + \sigma_g A^g + \kappa_l C^g + \sigma_{lg}(A^1_{lg} + A^2_{lg})$
P_l can be calculated from the difference

The general equation to calculate the expectation value of a measure X at a voltage V_{m} is

$$\langle X(V_{\text{m}})\rangle = \frac{1}{\mathcal{Q}(V_{\text{m}})} \sum_{l=1}^{N_r} \sum_{\mathbf{k}} n(\mathbf{k}) B(\mathbf{z_k}, r_l, V_{\text{m}}) X(\mathbf{z_k}, r_l, V_{\text{m}}).$$

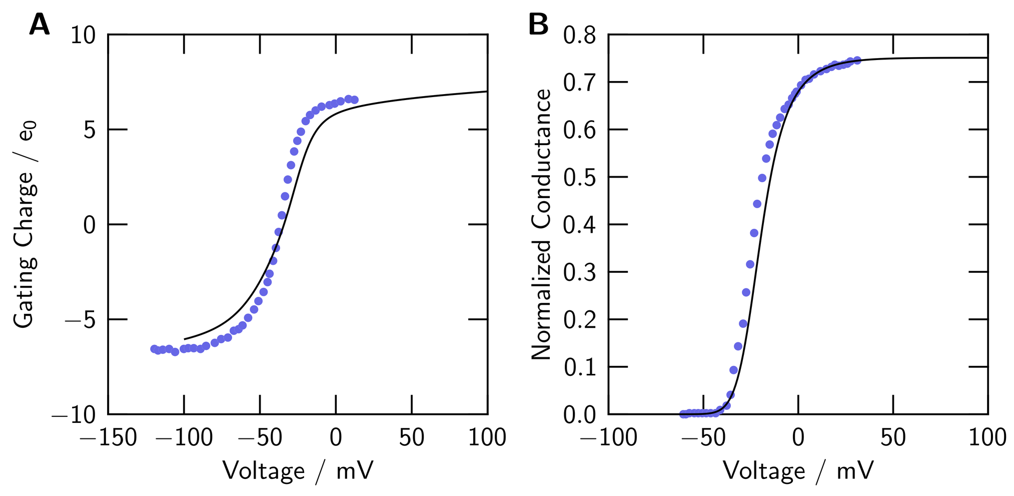
where $n(\mathbf{k})$ is the degeneracy of state $\mathbf{k}$

Work for operating the hydrophobic gate



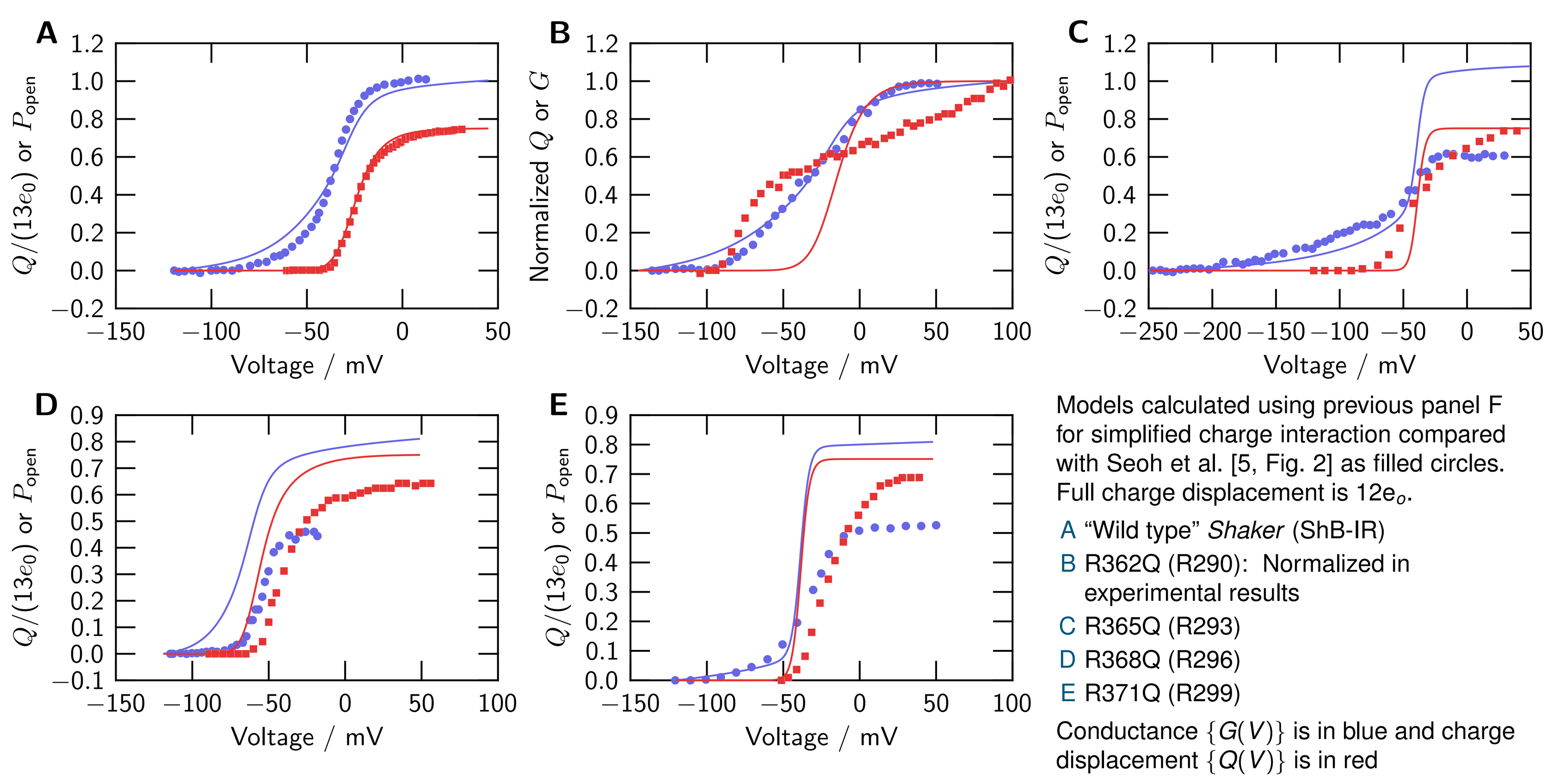
The work for operating the hydrophobic gate (*dashed line*) is small compared to the electrical work picked up by the four VS domains (*solid line*). The work for the VS is the expectation value $\langle\mathcal{H}_{\text{G}}\rangle$, and the electrical work is computed as $-\langle Q\rangle V_{\text{m}}$ where $\langle Q\rangle$ is the expectation value of the total gating charge displaced per channel [2]. Gate work is here referenced to that done at 0 mV.

Comparison of the full-channel model with experiment



(A) Gating-charge/voltage (Q/V) relation of the full-channel model (*solid line*); *symbols*: experimental Q/V relation of *Shaker* (ShB-IR) potassium channels (from Seoh et al. [5, Fig. 2A]); (B) Conductance/voltage (G/V) relation of the model (*solid line*), and the experimental G/V relation from Seoh et al. [5, Fig. 2A] (*symbols*).

Open probability and conductance: Hypothetical external charge as linear field



Models calculated using previous panel F for simplified charge interaction compared with Seoh et al. [5, Fig. 2] as filled circles. Full charge displacement is $12e_0$.

- A "Wild type" *Shaker* (ShB-IR)
- B R362Q (R290): Normalized in experimental results
- C R365Q (R293)
- D R368Q (R296)
- E R371Q (R299)

Conductance $\{G(V)\}$ is in blue and charge displacement $\{Q(V)\}$ is in red

Acknowledgments

This model was developed as a collaboration on the wild type *Shaker*-channel with Dirk Gillespie of the Department of Molecular Biophysics and Physiology, Rush University Medical Center, Chicago Illinois USA 60612 and Roland Roth of the Institut für Theoretische Physik, Eberhard Karls Universität Tübingen, 72076 Tübingen, Germany.

Partially funded by the Helmholtz Association through the Helmholtz Portfolio Theme "Supercomputing and Modeling for the Human Brain" and by National Institutes of Health grant GM083161.

Bibliography

[1] Alexander Peyser, Dirk Gillespie, Roland Roth, and Wolfgang Nonner. Domain and inter-domain energetics underlying gating in Shaker-type K_v channels. *Accepted: Biophys J*, 2014. doi: 10.1016/j.bpj.2014.08.015.

[2] Alexander Peyser and Wolfgang Nonner. Voltage sensing in ion channels: Mesoscale simulations of biological devices. *Phys Rev E Stat Nonlin Soft Matter Phys*, 86: 011910, Jul 2012. doi: 10.1103/PhysRevE.86.011910. URL http://dx.doi.org/10.1103/PhysRevE.86.011910.

[3] Alexander Peyser and Wolfgang Nonner. The sliding-helix voltage sensor: mesoscale views of a robust structure-function relationship. *Eur Biophys J*, 41:705–721, 2012. ISSN 0175-7571. doi: 10.1007/s00249-012-0847-z. URL http://dx.doi.org/10.1007/s00249-012-0847-z.

[4] Roland Roth, Dirk Gillespie, Wolfgang Nonner, and Robert E. Eisenberg. Bubbles, gating, and anesthetics in ion channels. *Biophys J*, 94(11):4282–4298, 2008. ISSN 0006-3495. doi: 10.1529/biophysj.107.120493. URL http://www.sciencedirect.com/science/article/pii/S0006349508700845.

[5] Sang-Ah Seoh, Daniel Sigg, Diane M. Papazian, and Francisco Bezanilla. Voltage-sensing residues in the S2 and S4 segments of the *Shaker* K^+ channel. *Neuron*, 16 (6):1159–1167, 1 June 1996. ISSN 0896-6273. doi: 10.1016/S0896-6273(00)80142-7. URL http://dx.doi.org/10.1016/S0896-6273(00)80142-7.

[6] Stephen B. Long, Xiao Tao, Ernest B. Campbell, and Roderick MacKinnon. Atomic structure of a voltage-dependent K^+ channel in a lipid membrane-like environment. *Nature*, 450(7168):376–382, 2007. doi: 10.1038/nature06265. URL http://dx.doi.org/10.1038/nature06265.