Chapter 2 - Supplementary information

1 Simulations of CRD of K_V7.2 channel

Molecular dynamics simulations of helices hA, TW, and hB were performed for three different configurations, wild type, introducing the mutation W344R and WT but starting from a tilted conformation; each situation was simulated following the same workflow and can be respectively found in the folders: **MUT WT WT_tilted**

All molecular dynamics simulations were performed with the software NAMD 2.13 [1] and the CHARMM36 all-atom force field [2]. The input structures were built from PDB 6FEG [3] by eliminating the N-terminal residual amino acids up to residue number 328, the W344R mutation was introduced by means of the visualization software VMD [4] and the conformation of the wild type starting from a tilted configuration was selected from the output of the previously introduced rosetta calculations,simulations were carried out in periodic cubic boxes of TIP3P water [5] so that the minimum distance of any protein atom and the edge of its box was at least 6.1 Å. To mimic neuron physiology, concentrations of 120 mM of KCl and 5 mM of NaCl were introduced. SHAKE bond length constraints were applied to all bonds, nonbonded interactions were calculated by the particle-mesh Ewald method with a cutoff of 12 Å for separating the long range and short range interactions.

The three folders contain the same information for each simulation, which is the following:

***./*:** parent folder of the simulation with coordinate (.pdb) and topology files (.psf).

step1_min_nvt: Minimization of the input structure using 1000 steepest descent steps followed by a NVT thermalization at 298K, keeping the temperature using Langevin dynamics with a Langevin damping of $0.5\,ps^{-1}$ [6] and a time step of 1 fs, for 0.5 ns.

- → 'emint1_hAB_mut.conf': input file for the minimization and first step of thermalization.
- → 'emint1_hAB_mut.restart.coor': binary file with the coordinates at the end of the first thermalization step.

step2_npt: NPT thermalization at 298K, controlling the temperature using Langevin dynamics with a Langevin damping of $0.5\,ps^{-1}$ [6] and the pressure with a langevin piston with a target pressure of 1 atm [7], for 1.5 ns.

- → 'th2_hAB_mut_NPT.conf': input files of the second step of thermalization.
- → 'th2_hAB_mut_NPT.restart.coor': binary file with the coordinates at the end of the second thermalization step.

step3_prod: Production with the same parameters as the previous NPT step but for 100 ns. This last step was repeated three times to obtain three different replicas, each of 100 ns of duration.

- → 'hAB_mut_long_run.conf': input run for the production run.
- → **replica_1**: results of the first production replica.

- ⋆ 'angles.png': time series of the angle of the run, angle definition can be found on the main text, chapter 3, Fig. 3.3.
- ⋆ 'rmsf.png': root mean square fluctuation of each amino acid on the sequence.
- ⋆ 'rms.png': total root mean square deviation of the sequence.
- ⋆ 'hAB_mut_long_run.restart.coor': binary file with the coordinates at the end of the production.

→ **replica_2**: results of the second production replica, same as replica 1.

→ **replica_3**: results of the third production replica, same as replica 1.

References

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