Structural mechanisms of gating in ionotropic glutamate receptors

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Movie Legends

Movies 1-9. Structural rearrangements in iGluRs during gating

Every movie starts and ends with the side view of the receptor parallel to the membrane. The GluA2 subunits A and C are colored purple, with B and D green. Molecules of the agonist glutamate (A) are represented by space-filling models. In some of the movies, Cα atoms for select labeled positions are shown as pink or cyan spheres. Movies are morphs between GluA2 structures representing the following states in the kinetic model shown in the bottom left corner: closed (C, GluA2-GSG1L$_{ZK-1}$, PDB ID: 5WEK), pre-active (P, GluA2$_{NOW}$, PDB ID: 4U4F), open (O with '*' for the conducting state, GluA2-STZ$_{Glu-CTZ}$, PDB ID: 5WEO) and desensitized (D, GluA2-2xGSG1L$_{Quis}$, PDB ID: 5VHZ).

Movie 1. Structural rearrangements in a single GluA2 LBD during gating

During pre-activation, binding of glutamate leads to ~11° rotation of the D2 lobe of LBD towards D1. LBD clamshell closure is much stronger (~26°) during receptor opening or desensitization.

Movie 2. Structural rearrangements in a GluA2 LBD dimer during gating

During pre-activation and opening, the upper D1-D1 interface stays intact, while glutamate binding leads to separation of the lower lobes D2 by ~4 Å during pre-activation and by ~13 Å during opening. During desensitization, the upper D1 lobes become separated by ~8 Å. The rupture of the D1-D1 interface in the desensitized state allows bringing the D2 lobes ~3, 7 and 16 Å closer to each other compared to the closed, pre-active and open states, respectively.

Movie 3. Structural rearrangements in the GluA2 LBD tetramer during gating

The LBD tetramer is viewed intracellularly along the receptor overall two-fold rotational symmetry axis. During opening, the LBD tetramer undergoes significant expansion (red arrows), with the maximum separation of the parts that are connected to the ion channel by LBD-TMD linkers. During desensitization, the A and C subunit LBDs rotate ~14° away from the subunit B and D LBDs. This rearrangement is accompanied by individual LBD dimers losing their local two-fold rotational symmetry, signified by a more four-fold symmetrical appearance of the LBD tetramer and pronounced clefts between protomers in each LBD dimer (blue arrows).

Movie 4. Structural rearrangements in GluA2 subunits A and C LBD-TMD linkers during gating

During opening, the M3 helices in subunits A and C become one helical turn shorter (orange arrows). Strong conformational changes are also observed in the S2-M4 linker, where the entire pre-M4 helix unwinds (green arrows), allowing extension of the S2-M4 linkers towards the ion channel pore.

Movie 5. Structural rearrangements in GluA2 subunits B and D LBD-TMD linkers during gating

During opening, the M3 helices in subunits B and D kink at the alanine A618 gating hinge (blue asterisk), leading to dramatic rearrangements in the ion conduction pathway.

Movie 6. Structural rearrangements in the GluA2 ion channel during gating

The ion channel is viewed extracellularly along the receptor's overall two-fold rotational symmetry axis. During opening, the M3 helices in subunits A and C become one helical turn shorter (orange arrows), the M3 helices in subunits B and D kink at the A618 gating hinge (blue asterisks) and the pre-M4 helices unwind (green arrows), allowing the S2-M4 linkers extension towards the ion channel pore.

Movie 7. Structural rearrangements in the GluA2 LBD tetramer top portion during gating

The LBD tetramer and ion channel are viewed extracellularly along the overall two-fold rotational symmetry axis. During opening, the LBD tetramer undergoes significant expansion (red arrows), leading to a slight clockwise rotation of the entire LBD tetramer layer. During desensitization, the ~14° rotation of the A and C subunit LBDs away from the subunit B and D LBDs results in a substantial overall clockwise rotation of the entire LBD tetramer layer.

Movie 8. Structural rearrangements in the GluA2 ATD tetramer during gating

The GluA2 tetramer is viewed extracellularly along the overall two-fold rotational symmetry axis. During gating, the ATD tetramer moves as a rigid body, rotating clockwise by ~8° upon opening and ~18° upon desensitization.

Movie 9. Overall movements of GluA2 during gating

The GluA2 tetramer is viewed parallel to the membrane. During gating, the entire receptor undergoes twisting, with the strongest corkscrew motion observed upon desensitization, when the receptor becomes ~5 Å shorter than in the pre-active or desensitized states.