Cholesterol Dependence on the Conformational Changes of Metabotropic Glutamate Receptor 1 (mGluR1)

Ugochi Isu, Adithya Polasa, Seyed Hamid Tabari, Mortaza Derakhshani-Molayousefi, Mahmoud Moradi
Chemistry and Biochemistry, University of Arkansas, Fayetteville, AR, USA

INTRODUCTION

- Metabotropic glutamate receptors (mGluRs) are class C G-protein-coupled receptors (GPCRs) which exist as constitutive dimers.
- They play significant roles in regulating neurotransmission and activating excitatory synapses in the central nervous system.
- We have used all-atom molecular dynamics simulation to show the interaction of cholesterol on the local structural dynamics of mGluR1 7TM domain in an inactive model. Our simulation characterizes the conformational changes of mGluR1 in 0%, 10% and 25% cholesterol concentrated lipids.
- Our results reveal that cholesterol influences the conformational changes of the internal protein and acts less significantly on individual protomers.
- Our analysis show that low cholesterol (10% cholesterol: 90% POPC) induces more significant conformational changes in mGluR1, while the system with higher cholesterol (25% cholesterol: 75% POPC) tends to behave similarly to systems without cholesterol (0% cholesterol: 100% POPC.
- Due to high sequence conservation of the TMDs of mGluRs, the molecular interactions we have observed showing cholesterol dependence in mGluR1 are likely to be applicable for other members of the mGluR family.

METHODS

We have investigated the influence of cholesterol on mGluR1 dynamics. The crystal structure of Human class C mGluR1(PDB entry: 4OR2) in complex with a negative allosteric modulator was obtained from Protein Data Bank (PDB) as initial structure. Cholesterol molecules bound between the monomers were removed. The proteins were placed in homogenous and heterogenous membrane lipids, solvated in a box of TIP3P waters, and 0.15M NaCl. The heterogenous lipids consisted of 10% cholesterol/90% POPC and 25% cholesterol/75% POPC with total atom size of 196337 and 192820 respectively, while the system with homogenous lipid comprised of pure POPC with 152713 atoms in total. All three (3) systems were run for 1 microsecond each. The root mean square deviation (RMSD) trajectory tool of VMD was used to calculate the RMSD

CONCLUSION

- Our results indicate that the influence of cholesterol on mGluR1 shows higher significant conformational changes in whole protein than in singular protomers.
- We also show that cholesterol in mGluR1 is localized more in the interphase of the protein..
- We observe that the system with 10\% cholesterol tends to show higher significant changes than at 25\% cholesterol.
- This could suggest that as cholesterol concentration increases, the protein becomes more ordered hence less motion between the helices.

RESULTS

1. INTERACTION SITES OF CHOLESTEROL IN MGLUR1

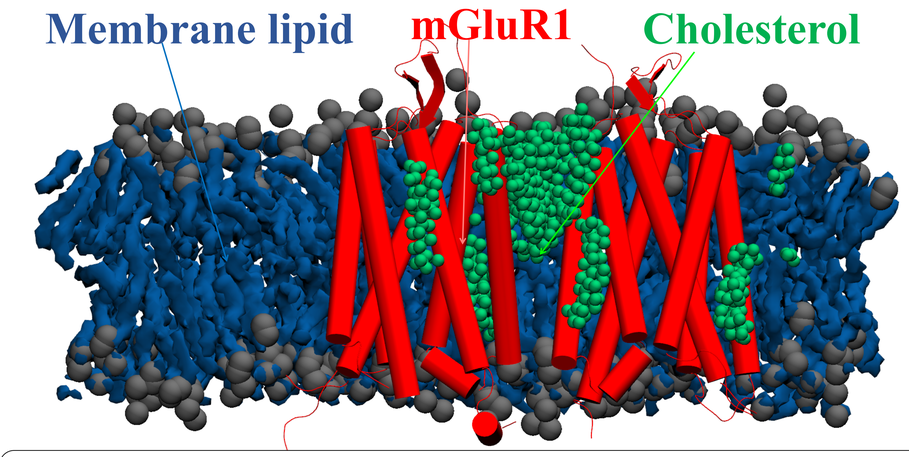

Figure 1. Cholesterol molecules (shown in green, with head groups shown in red) interacts between the monomers of mGluR1 (shown in gray) and between the groves of the transmembrane helices

2. IONIC INTERACTIONS BETWEEN TRANSMEMBRANE DOMAINS SHOW THAT LOW CHOLESTEROL CONCENTRATION AFFECTS THE DYNAMICS OF MGLUR1

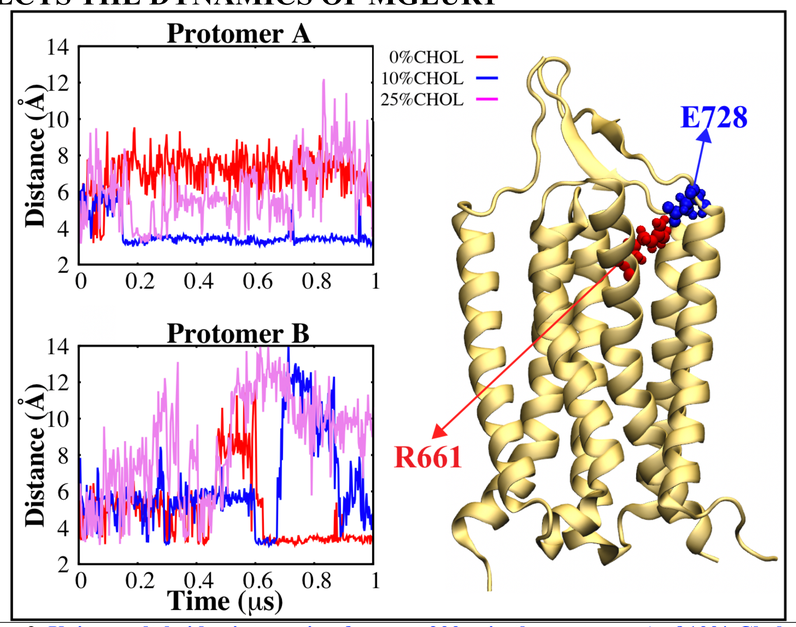

Figure 2. Unique salt-bridge interaction forms at 200ns in the protomer A of 10% Cholesterol, and remains stable throughout the simulation. **Figure 2** shows the time series of R661–E728 salt-bridge distance and the visual representation of salt-bridge formation in the protomer A of 10% Cholesterol

3. CHOLESTEROL INFLUENCES THE CONFORMATIONAL CHANGES OF THE INTERNAL PROTEIN AND ACTS LESS SIGNIFICANTLY ON INDIVIDUAL PROTOMERS

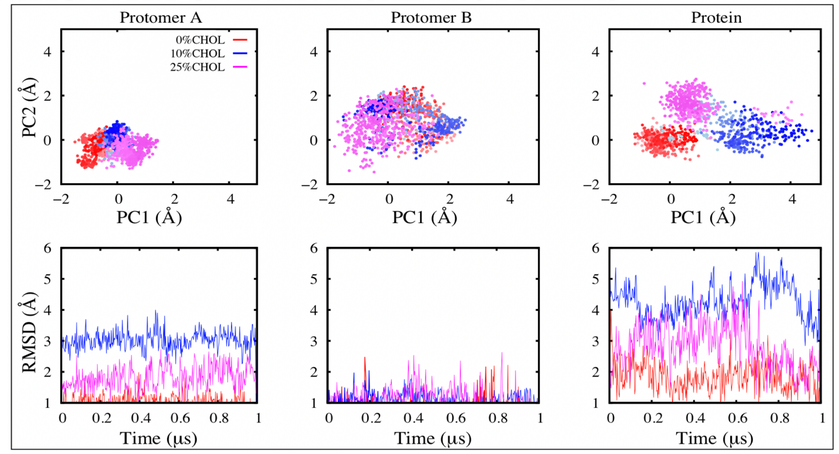

Figure 3. Projections of the principal components (PC's) 1 and 2 [A-C], and the root mean square deviation analysis [D-F] of mGlUR1 in the presence and absence of cholesterol. We observe higher conformational differences in the whole protein than in individual protomers. Protomer A revealed more fluctuations when comparing only the protomers

4. LOW CHOLESTEROL CONCENTRATION INDUCES A HIGHER CONFORMATIONAL CHANGE AS OPPOSED TO THE SYSTEM WITH HIGH OR NO CHOLESTEROL

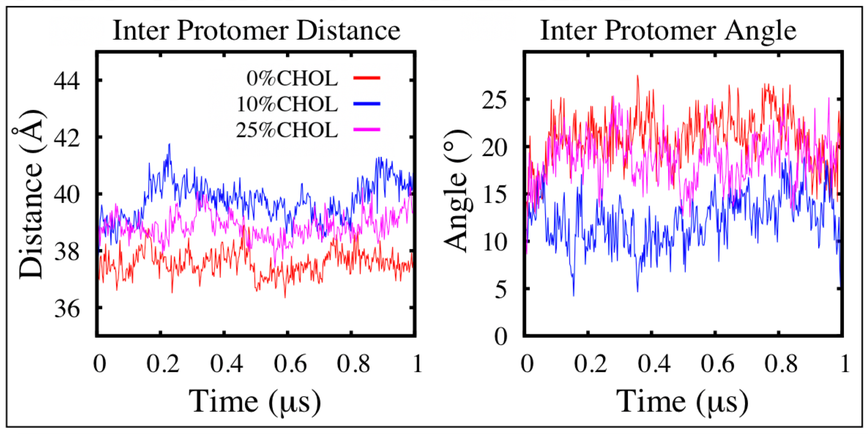

Figure 4. Time series representation of the inter protomer distance (left panel) and inter protomer angle (right panel). As the inter protomer distance increases in 10% cholesterol, the interprotomer angle decreases

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