Rhodopsin Dimers: Molecular Dynamics Simulations Using Discrete Representations of the Membrane and Water Environment

Simon X. Wang, Marta Filizola, Marc Ceruso, Harel Weinstein

Department of Physiology and Biophysics Weill Medical College of Cornell University, New York, New York 10021

INTRODUCTION

G protein-coupled receptors (GPCRs) comprise by far the largest family of cell surface proteins involved in signaling across the plasma membrane and are implicated in numerous diseases. Direct evidences for homo- and/or hetero-dimerization of various GPCRs reinforces the need for the incorporation of these key phenomena into any physiologically relevant functional models of these receptors, especially in light of the recently discovered functional implications of GPCR association, which include pharmacological diversity, G-protein coupling, downstream signaling, and internalization.

Both computational and experimental efforts have been made to understand the basis of protein-protein interaction in GPCR dimerization. Our computational approaches based on correlated mutation method and three-dimensional molecular models of the transmembrane segments of GPCRs elucidated the likely molecular determinants required for dimerization [1, 2]. Recent experimental data coming from an atomic-force microscopy (AFM) map of rhodopsin molecules in their native mouse disk membranes, support a molecular model of rhodopsin monomers organized into two dimensional arrays of dimers [3]. Specifically, transmembrane (TM) helices 4 and 5 of rhodopsin appear to be involved in intramolecular contact, whereas helices TM1 and TM2, and the cytoplasmic loop connecting helices TM5 and TM6 are inferred to facilitate the formation of rhodopsin dimer rows.

Since the nature and geometry of the interface of GPCR dimers is essential to the understanding of the role and implications of these phenomena in the functional mechanisms of these receptors, we take advantage of the information available for rhodopsin to carry out a computational study that could provide detailed insights into the determinants of GPCR dimerization in a structural context. As a first step, molecular dynamics (MD) simulations are carried out for the systems for which direct structural information is available, using discrete representations of the membrane and water environment.

COMPUTATIONAL METHODS

Simulation Setup All simulations are performed for an NPT ensemble using GROMACS 3.2.1 package. Lipid parameters were taken from Berger et al. [6] and the water model was SPC. The system was simulated with semiisotropic coupling, with the pressure at 1.0 bar, coupled separately to the x and y directions. The Particle Mesh Ewald interpolation order was set to 6 and the maximum grid spacing for the FFT was set to 0.12 nm.

Helix Analysis New algorithms were developed [5] for the analysis of helix properties and their evolution in the dynamics simulations. (Simulated http://fulcrum.physbio.mssm.edu/~mezei/simulaid)

RESULTS

The current trajectories length is 20 ns for both monomer and dimer. The Left Panel above records the rms deviation of TM bundle of rhodopsin monomer, subunit A and B and whole dimer from the crystallographic structure. Comparison to the crystallographic B-factors (Right Panel) shows that the overall trends of the C-alpha atom fluctuations in a structural context. As a first step, molecular dynamics (MD) simulations are carried out at the systems for which direct structural information is available for rhodopsin to carry out a computational study. The line represents movement of center of mass of subunit A and B. Preliminary results from the 20 ns trajectories of rhodopsin dimer and monomer suggest subtle structural changes at the dimer interface. Subtle differences in some of the calculated helix properties were observed thus far for i) the helix rotation of TM4, TM5, TM7, and HR around their own axes; ii) the displacement of TM5, TM4, and HR in the z and/or y direction.

CONCLUSION

Preliminary results from the 20 ns trajectories of rhodopsin dimer and monomer suggest subtle structural changes at the dimer interface. Subtle changes were also observed at distant domains that are critical to receptor activation. Control simulations of various lengths and using different equilibration protocols are ongoing to assess the accuracy of simulations and of the inferences regarding conformational changes.

REFERENCE

5. Mezei, M., Filizola, M. manuscript in preparation.