Top-down Multiscale Models and Free Energy Calculations of Multivalent Protein-Protein and Protein-Membrane Interactions in Nanocarrier Adhesion and Receptor Trafficking

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Abstract:

We present recent development and applications of top-down mesoscale modeling to two biologically relevant problems [1]: (1) adhesion of nanocarriers to cells mediated by multivalent receptor-ligand interactions in targeted drug delivery; (2) internalization of cell surface receptors in cells via the biological process of endocytosis. In particular, we focus on methods for computing absolute/relative free energies using these mesoscale models in order to facilitate direct comparison with experimental data. Experimental technologies such as x-ray crystallography and nuclear magnetic resonance are well established at the atomic resolution (1-10 Å) and optical microscopy methods are well established at the micron resolution. However the mesoscale resolution (1-200 nm), which is the most relevant for functional signaling modules, has been elusive. Multiscale modeling complements experimental technologies in order to access the mesoscale (10-100nm). Traditional multiscale modeling involves bottom-up approaches of systematically coarse-graining the atomistic description, in order to access the mesoscale. Here, we describe a top-down approach, in which models are constructed at the mesoscale based on phenomenological interaction potentials, and the parameters are determined directly by independent biophysical experimentation. In prior work published in the literature, such an approach has been employed, and specific choices of the governing equations have been validated based on experimental studies. While the top-down strategy is already proven to be a viable avenue for pursuing models that provide physical insight, new computational methodologies are required to enable direct comparison with experiments. In this work, we illustrate such methodologies through the use of free energy calculations and illustrate the power of our approach on two biologically relevant problems: (1) adhesion of nanocarriers to cells mediated by multivalent receptor-ligand interactions in targeted drug delivery; (2) internalization of cell surface receptors in cells via the biological process of endocytosis.

Targeted drug delivery using functionalized nanocarriers (i.e. carriers coated with specific targeting ligands) represents a promising approach in therapeutic applications. However, targeting of nanocarriers (NCs) to endothelial cells (ECs) remains an important design challenge in biomedical science. The use of functionalized NCs offers a range of tunable design parameters (i.e., size, shape, etc.) and a high-dimensional tunable parameter space needs to be spanned to determine optimal design. Challenges inherent to design include: (i) molecular and geometric parameters surrounding receptor-ligand interactions and NCs, (ii) accurate characterization of hydrodynamics, (iii) physico-chemical barriers for NC uptake/arrest/internalization, and (iv) uncertainty in targeting environment, to name a few. Binding affinity or association constant is a well defined physico-chemical measure of the efficiency of NC targeting. This quantity has been experimentally measured and often employed in guiding the rational design of functionalized NCs. Despite such previous studies on NC binding, a comprehensive understanding of the determinants of NC binding to EC is still limited. The challenge, from a modeling perspective and one we address by devising a strategy to compute the absolute free energy of binding, is to predict how the binding affinity depends on experimentally tunable parameters; such a

computational frame-work, as we illustrate, is very powerful in its ability to aid in the optimization of NC design [2, 3].

Curvature-driven processes in cell membranes are of considerable interest to intracellular trafficking, organelle homeostasis, and biogenesis. Several recently discovered protein membrane-binding domains have been postulated to assemble in a process that is driven by membrane curvature and membrane tension and in the process induce local deformations of the bilayer. During the process of endocytosis, clustering of proteins with Epsin N-terminal homology (ENTH) domain in regions of background mean curvature have been reported. The mechanism by which clathrin sustains curvature in the bud region is another open question in clathrin-mediated endocytosis (CME). The emerging view is that membrane tubulating proteins (e.g. epsins) are incorporated as part of the growing coat through clathrin or adaptor protein 2 (AP-2) mediated interactions mediated by the clathrin/AP-2 binding region (CLAP) domain on epsin, in order to stabilize the membrane curvature. Efforts to determine the interaction map of CME-related proteins using established methods in structural biology and biochemistry have yielded successful results. Yet, crucial to our understanding of endocytosis, and still elusive, is the nature of spatial and temporal localization of these proteins. Through a mesoscale model along with mean-field energetics as well as thermodynamic integration, we provide insights at the 1-200 nm resolution, which aid in our understanding of the structural and energetic aspects of curvature driven forces and vesicle assembly in the cellular process of endocytosis. In particular, our study yields a minimal model for signaling and trafficking for CME, which is simultaneously consistent with experiments on structure, energetics, and signaling dynamics [4,5].

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Keywords: Targeted drug delivery, Clathrin-mediated endocytosis, Monte Carlo, Helfrich free energy, Umbrella sampling, Absolute binding free energy, Thermodynamic integration

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