Equilibrium dynamics in protein crystals

The mechanical properties of proteins allow them to visit multiple conformational states while carrying out biological functions. In order to understand, predict, and control biological processes at the molecular level, it is essential to precisely characterize protein structural dynamics at the atomistic scale. New “pump-probe” methods such as electric-field stimulated time-resolved X-ray diffraction (EFX) open up ways to initiate and observe protein motions with atomistic detail on biologically relevant timescales. However, the complexity of these experiments demands the parallel development of effective molecular dynamics approaches to accelerate progress and extract meaning. Here, we begin this work by establishing robust and accurate methods for simulating equilibrium dynamics in protein crystals. Applied to the second PDZ domain of the human LNX2 protein, our study reached three primary objectives. Firstly, we determined the optimal simulation setup, including force field and environmental composition, for modeling the experimentally observed equilibrium state of the PDZ domain. Secondly, we explored the conformational ensemble sampled from various simulation setups to define the key factors affecting the model accuracy. Finally, we estimated the agreement with the experimental data across multiple independent metrics, such as protein structure, flexibility, crystallographic symmetry, intermolecular interactions and crystallographic water. We addressed the functional relevance of mechanical deformations induced by thermal fluctuations at equilibrium. This work lays the foundation for a virtuous cycle between simulation and the new pump-probe experiments for visualizing and understanding protein reaction coordinates.