Molecular Simulations of Alternate Frame Folding in an Engineered Ca2+-Sensing Protein Switch

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A promising strategy for engineering protein conformational switches is the “alternate frame folding” design in which a protein is fused in an end-to-end fashion to a circular permutant of the protein such that the two proteins partially overlap in sequence. Due to the sequence overlap, the folding of the proteins is mutually exclusive. Here we characterized the mechanism of alternate frame folding in an engineered Ca^{2+}-sensing protein switch using molecular simulation. The simulations were carried out using the weighted ensemble path sampling strategy, which increases the efficiency of sampling rare events (e.g., large conformational transitions) without introducing any bias into the dynamics. Based on these simulations, we have made predictions of mutations that may improve the kinetics of switching while maintaining the signal response of the switch. Experimental validation of our simulations will also be presented.