FIAS Frankfurt Institute for Advanced Studies

CMMS

Talks

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Probing the hidden dynamics and energetics of diverse biomolecular systems by AFM

The forces and energetics that stabilize membrane proteins remain elusive to precise quantification. Single-molecule force spectroscopy can yield kinetic rate constants, energetics, intermediate states, unfolding pathways, and even a projection of the underlying free-energy landscape. Using recently developed micromachined AFM cantilevers, we reexamined the unfolding of individual molecules of bacteriorhodopsin (bR) embedded in its native lipid bilayer with a 100-fold improvement in time resolution and a 10-fold improvement in force precision. Numerous newly detected intermediates—many separated by as few as 2–3 amino acids—exhibited complex dynamics, including frequent refolding and state occupancies of <10 μs. To quantifying energetics, we leveraged the rapid and reversible initial unfolding of bR to determine the free-energy change ($\Delta\Delta G0$) for select point mutants and thereby highlighted the importance of measuring membrane protein energetics in lipid bilayers rather than mixed micelles. In bR's photocycle, photon absorption by retinal triggers a conformational cascade that results in pumping a proton across the cell membrane. By integrating sub-ms illumination with our AFM, we quantified the energetics of a light-induced change in bR's photocycle. I will conclude by discussing extensions of these enhancements in bioAFM to other diverse biomolecules, including nucleic-acid structures and globular proteins involved in pathogenesis.

When: Tuesday, 12 November 2024, 11:00
Where: FIAS Lecture Hall (0.100)
Ruth-Moufang-Str. 1, Campus Riedberg