MS08 Serial crystallography, obtaining structures from many crystals

MS8-03
Molecular snapshots of drug release from tubulin
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Abstract
The dynamic interplay between proteins and their ligands is central to molecular biology, pharmacology, and drug development. Simple static models of ligand-protein interactions such as the “lock-and-key” mechanism have long been known to be insufficient. Much needed molecular insight can be obtained by resolving structural intermediates, but the relevant transitions extend over many orders of magnitude in time and cannot be resolved with conventional structural biology.
We have used time-resolved serial crystallography at a synchrotron and X-ray laser, to study the release of the photochemical affinity switch azo-Combretastatin A4 from the anti-cancer target tubulin. Thirteen logarithmically spaced temporal snapshots at near-atomic resolution are complemented by time-resolved spectroscopy and molecular dynamics simulations. They show how the photoinduced cis to trans isomerization of the azobenzene bond stretches the ligand in the picoseconds, the formation of a metastable binding pose in the nanosecond range which is followed by stepwise opening of a gating loop within microseconds, and the completion of the unbinding reaction within milliseconds. Ligand unbinding is accompanied by a collapsing binding pocket and global tubulin-backbone rearrangements. Our results have implications for the molecular basis of photopharmacology, the mechanism of action of anti-tubulin drugs and provide an experimental framework to study protein-ligand interaction dynamics.