LECTURE SERIES:

Interfaces of Chemistry, Life Sciences and Physics

Friday, November 14, 2014
At 3:30 p.m.
172 St. John's College (Schultz Theatre)

Wine and Cheese Reception to Follow

Enzymatic Transition States, Analogues and Dynamics in Catalysis:

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Abstract

Knowledge of transition state structure for enzymatic reactions permits the design and synthesis of transition state analogues. In thermodynamic theory, binding of transition state analogues is proportional to the catalytic rate enhancement provided by the enzyme. Since enzymes can enhance rates up to 10^{21} – fold, well designed transition state analogues are expected to bind more than a million times tighter than their substrates. Some transition state analogues can serve as drug candidates. Our approach uses multiple intrinsic kinetic isotope effects and computational chemistry to solve the structure of enzymatic transition states. An electrostatic potential map of the transition state formed on the enzyme is used as a target for the design and synthesis of chemically stable transition state analogues. The procedure for targeting a specific enzyme for transition state analogue design involves: 1) selection of the target enzyme, 2) synthesis of isotopically labeled reactant molecules, 3) measurement of intrinsic kinetic isotope effects, 4) match isotope effects with quantum chemistry models of the transition state, 5) design analogues with similarity in geometry and electrostatic potential to the transition state, 6) chemical synthesis of transition state analogues, and 7) testing transition state analogues against the targeted enzyme in enzymatic assays, cell and animal models.

Transition state structure based on kinetic isotope effects permits the design of powerful transition state analogue inhibitors, even for closely related enzymes. Inhibitors of various enzyme targets show promise in T-cell disease, gout, malaria, ulcers and cancer. Transition state analogues provide chemical probes to explore protein structure and dynamics. Transition states and transition state analogues are opposites on the chemical time scale. Transition states are formed by femtosecond (fsec) coupled motion through stochastic local dynamic searches at the catalytic site. Transition state analogues trap thermodynamic energy and are released slowly (min to hr) from enzymes. Transition state lifetime in the example of human purine nucleoside phosphorylase (PNP) is 10^{-16} of the release rate for a transition state analogue. Computational and experimental chemical clocks indicate that local catalytic site motions on the fsec time scale modulate transition state formation in PNP. Fsec protein motions in PNP can be perturbed in human PNP by replacement of H, C and N with ^{2}H, ^{13}C and ^{15}N to generate an enzyme with altered bond vibrational structure. Heavy PNP shows changes in the on-enzyme chemistry rate but steady-state kinetics and transition state structure are unchanged. Mass-perturbation of local modes decreases the frequency of local motions involved in the transition state. Heavy proteins offer a new tool for investigation of protein motion in catalysis.

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