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Vern L. Schramm was born in South Dakota and earned his Bachelor's degree from South Dakota State College. He obtained a Master's degree in nutrition from Harvard University and a Ph.D. in the mechanism of enzyme action, studying with John F. Morrison at the Australian National University. After two years as a NSF–NRC postdoctoral fellow at the NASA Ames Research Center, he joined the faculty of Temple University School of Medicine, where he spent 16 years. In 1987, he moved to the Bronx to become Professor and Chair of Biochemistry at the Albert Einstein College of Medicine. He was named the Ruth Merns Chair of Biochemistry in 1995. Honors in recognition of his contributions include a Merit Award from the NIH, the Rudi Lemberg Award from the Australian Academy of Science, and the Repligen Award for 2006 from the Biological Chemistry Division of the American Chemical Society. Research in the Schramm laboratory focuses on enzymatic catalysis, transition state structure, dynamics in catalysis, and molecular distortion in enzyme–ligand interactions. Transition state analogue inhibitors from this approach are now in clinical trials.

Introduction: Principles of Enzymatic Catalysis

The catalytic rate enhancement imposed by enzymes has attracted the attention of chemists for more than a century. Our understanding of enzymatic catalysis has evolved slowly, in parallel with the development of experimental tools to probe chemical and kinetic mechanisms. The relatively recent explosion of three-dimensional structures from crystallographic techniques has provided detailed insights into the architecture of enzymes and their interactions with reactants. Still, it has been difficult to fully account for the enzymatic power from these static structures. Today, it is increasingly clear that molecular dynamic contributions to catalysis must be incorporated to provide a full understanding of catalysis.

The first section of this issue of *Chemical Reviews* addresses experimental approaches being used to explore dynamic contributions to catalysis. Spectroscopic probes explore time-resolved dynamics of protein conformational changes and the influence of reactant interactions. Unlike our useful, long-standing, and beloved construct of single kinetically distinct species representing each step through the reaction coordinate, direct dynamic spectroscopic measurements indicate that multiple enzymatic conformational states exist even in kinetically simple Michaelis complexes. Likewise, the kinetic definition of the free enzyme as a single state is in reality a large number of protein conformational states. This protein disorder is clearly seen in the dynamic motion measured by distance- and time-resolved NMR studies. Single enzyme molecule reactions can now be examined by fluorescence and related spectroscopic techniques. A single enzyme molecule gives kinetic signals that reflect the dynamic states of individual catalyst molecules, information that is lost in the average signals from ensembles. Dynamic contributions to catalysis are also implicated in the measures of isotope effects that occur in hydride transfer reactions. Perturbations that influence dynamic motions of proteins also influence the transition state structures in a manner implicating dynamic contributions to the transition state. Hydride tunneling at the transition state is a sensitive manifestation of the impact of dynamics on chemistry. Molecular dynamics in enzymatic catalysis is an experimentally difficult field because of the extreme differences in the time scales for the chemical step and enzymatic turnover numbers. For example, transition states are theorized to exist within the time for conversion of a bond restoring mode to an atomic translational mode, identified with the imaginary frequency in transition state nomenclature. As the time of a single bond vibration is on the order of 10^{-13} s in the case of hydride transfer, enzymatic transition states have lifetimes that are resistant to direct spectroscopic probes for solution reactions. In contrast, most enzymes have catalytic turnover times near 10^{-2} s. The time scale disconnect can be

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interpreted to mean that enzymatic dynamic excursions are searching for as many as 10^{11} transition state lifetimes to generate one successful transit of the transition state barrier.

The opposite problem exists in computational chemistry. Bond vibrations of extended systems including enzyme and reactants can be computationally simulated with some accuracy for time periods from the femtosecond to the nanosecond time scale. This provides computational dynamic access to the time scale of the transition state, and when dynamic and quantum mechanical effects are coupled, interesting models of transition state chemistry result. However, the 10^{-2} s time scale commonly encountered in enzymatic catalysis is far beyond continuous dynamic calculations. A frontier in computational analysis is to find robust techniques to sample the long time scales of enzymatic catalytic cycles and to impose both dynamics and quantum chemistry over realistic time scales by sampling methods.

The second group of reviews provides perspective on catalysis from advances in computational enzymology. Traditional views of catalysis treated the transition state in thermodynamic terms, assuming an equilibrium between reactants and the transition state. With the introduction of dynamic contributions to catalysis, the thermodynamic notion of equilibration between reactants, the transition state(s), and possible covalent intermediates presents a challenge. These issues are addressed in an examination of transition state complexes as covalent intermediates and free energy considerations from computational dynamics and quantum chemical analyses. The problem of computational chemical approaches to span the extreme time scales from bond vibrations to enzymatic turnover rates is also explored. Finally in this section, the concepts of multidimensional tunneling and the transmission coefficient are examined for their role in enzymatic catalysis. As emphasized in one of these reviews, computational chemistry is entering the field of enzymatic catalysis as an essential tool for the exploration of every mechanism. Incorporation of new technologies to understand enzymatic catalysis is a recurring theme in enzymology. The application of kinetic measurements, kinetic isotope effects, crystallography, and NMR are past examples, and computational chemistry is developing rapidly.

The dynamic approaches in the first two sections provide a prelude to the central theme of catalysis, mechanisms of chemical activation through electron donation or withdrawal to lead to bond making and bond breaking. Nucleophilic reactions are common to many chemical reactions, and enzymology is no exception. Investigation of the nature of oxygen nucleophiles at or near enzymatic transition states offers a new perspective on this reaction class. Included in the chemistry of oxygen nucleophiles is the transfer of phosphate and sulfate groups. Precise isotope effects from oxygen have revealed the nature of transition states for nearly all classes of these transfers. Common catalytic site reactivity, for example, in the serine protease family, predicts covalent modifications of an enzymatic reaction class with class-specific site-directed chemistries. Systematic trapping of all catalytic sites containing this reactivity provides a catalytic site proteomic and genomic approach to protein catalytic function. Patterns of reactive enzymes as a function of tissue status are used to provide mechanism-based profiling for diagnostic insights.

A continuing development in enzymatic reaction mechanisms is the understanding of free radical chemistry at catalytic sites, which has advanced even since the field was reviewed in this Journal in 2003 (Vol. 103). Radical chemistry provides sufficient chemical potential for carbon–carbon rearrangements and other difficult reactions and requires radical-generating cofactors or cosubstrates. But most enzymes use bond polarizing effects to initiate bond changes. Metal ions bound at catalytic sites provide a chemical scaffold for bond polarization, and two review articles address this topic. In the first article, the use of common scaffolds for uncommon and difficult reactions, including some radical reactions, is explored. The second review article deals specifically with activation of water nucleophiles at catalytic sites using binuclear metallohydrolases. Halogenation reactions are rare in higher organisms but are of growing importance in bioremediation chemistry. Analysis of enzymes capable of halogenation reactions indicates that most reactions involve oxidative strategies. Finally, in this section, the chemical reactivity of water is compared in nonenzymatic and enzymatic reactions. These comparisons provide benchmarks for enzymatic catalytic proficiency and are valuable in estimating the energy available for catalysis and for conversion into binding energy for chemical analogues of enzymatic transition states.

Many enzymes catalyze unusually difficult reactions with high specificity. Enzymes that edit specific adenylate residues on large messenger RNA molecules show such specificity and lead to a change in the genetic code as a result of specific deaminations. The family of terpenoid cyclases catalyze reactions with extreme stereochemical control and substrate specificity. These reactions lead to the tens of thousands of terpenoids found in nature. DNA repair enzymes are also capable of extreme specificity, scanning large regions of normal base pairs with no activity until a rare mutation, differing in only one or two atoms, causes initiation of catalysis. As more catalytic mechanisms are revealed, we are reminded that enzymes evolve to function and often incorporate neighboring group effects for bond polarization. Participation by the electron-rich phosphoesters in nucleic acids and nucleotides provides examples of this effect in catalysis. The final review describes the genetic and enzymatic equivalent of an assembly line in the chemical synthesis of the thousands of polyketides and nonribosomal peptide antibiotics found throughout nature, thereby providing many of the most valuable antibiotics. The chemical mechanisms of these enzymes not only provide insight into the logic of complex synthetic pathways, but also hold promise for genetic alterations and manipulations of these pathways to create new antibiotics.

Any collection of reviews on a topic as broad as principles in enzymatic catalysis will always be incomplete. Limitations of space, author availability, and recent reviews published elsewhere are only a few of the factors that make this collection deficient in some areas. These deficiencies are the sole responsibility of the Guest Editor and not the participating authors, who have invested greatly of their time and talent. The result provides a timely compilation of new insights into catalysis, integrated into past knowledge, to inform and assist other researchers in the developing concepts of enzymatic catalytic function.

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