Catalytic Antibodies and Disfavored Reactions

Samuel Danishefsky

Most valuable chemical transformations (*1*) require either a promoter or a catalyst. The promoter unlike the catalyst may, in itself, be consumed in the reaction. Even when the promoter is not dissipated in the primary reaction, it tends to exhibit a relatively low turnover efficiency. Therefore the promoter, unlike the catalyst, will in general be used in stoichiometric (or even greater) proportions. By contrast, catalysts can be used in subequivalent quantities. Not surprisingly then, the search for new, accessible, and efficient catalysts lies at the center of chemistry.

In the quest for improved catalytic efficiency, it might seem reasonable to start with enzymes (*2*). The overwhelming majority of enzymatic functions are performed by proteins. Nature has brilliantly exploited the diversity inherent in the primary, secondary, and tertiary structure of proteins to foster catalysis of unparalleled efficiency and specificity that orchestrates virtually all of the strategic reactions needed for life. Little opportunity for gross improvements in efficiency, as judged by rate, seem to be available in the refashioning of enzymes (*3*).

With the efficiency and selectivity of the natural enzymes comes a difficulty for organic chemists who would seek to exploit them in general contexts. Having evolved apparently for the purpose of conducting specific, life-enabling reactions, the applications of enzymes to nonnatural situations have proven to be rather more complicated. It is appropriate to distinguish between two kinds of nonnatural settings. One is that of a natural type of reaction (such as oxidation, reduction, or aldol condensation) with nonnatural substrates. While impediments to substrate tolerance are encountered, considerable latitude in achieving enzymatic modulation of artificial substrates undergoing natural reactions has often been achieved (*4*). Much less tractable has been the goal of adapting protein-based enzymes to catalyze reaction types not included in their original "job description." Not surprisingly, the exquisitely tuned protein structure of enzymes has shown little adaptability (or imagination) in taking on wholly unanticipated assignments.

Two daunting complications stand in the way of constructing de novo protein-based catalysts (that is, artificial enzymes) to accommodate unnatural reactions. A highly detailed understanding would be necessary to interrelate the active site structure of the proposed protein with its prowess for catalysis. Moreover, even if an optimum desired three-dimensional disposition of the active site could be formulated with appropriate resolution, a high order of predictability in the relationship of primary structure and functionally folded unnatural protein would be required (*5*). For the moment, the capacity to obtain peptides and proteins of defined amino acid sequence by fully synthetic or recombinant means has not been of major consequence in obtaining valuable de novo fashioned artificial enzymes.

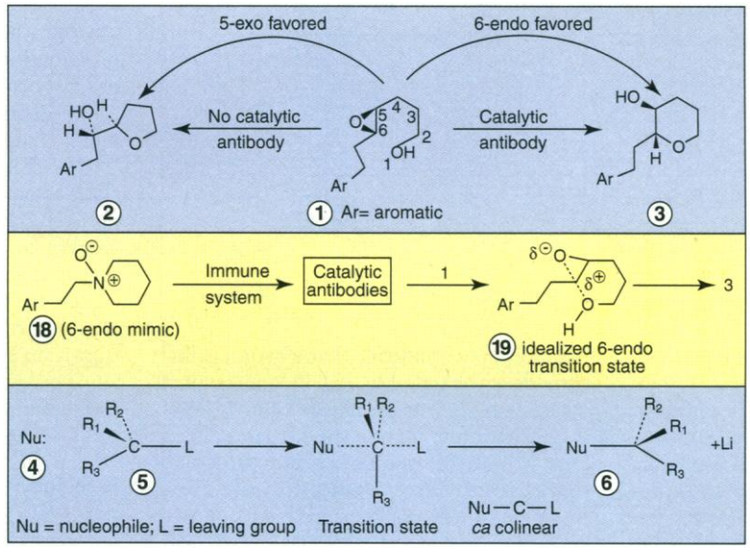
It is for this reason that catalytic antibodies have captured the fancy of many chemists. The field has been amply reviewed of late (*6*, *7*), and I will only offer the most general of observations. The massive power of the immune system is directed toward producing antibodies whose combining sites are complementary to antigen which is so designed as to simulate the proposed transition state for a chemical reaction. When exposed to reaction substrates, the binding forces of the antibody cause acceleration of reaction, by prompting their reactants to take on the character of the transition state required for a chemical transformation. Given a proper turnover rate and given ideal hapten selection, virtually the full binding force of the antibody is, in principle, applicable for purposes of catalysis (*8*). The capacity to generate and tap catalytic antibodies to catalyze known but nonnatural chemical reactions has been well demonstrated (*6*, *7*).

Hitherto, catalytic antibodies have been used to catalyze reactions that fall comfortably in the purview of "standard" organic chemistry (including, among other processes, ester hydrolysis, trityl hydrolysis, amide bond formation, Diels Alder reaction, Claisen rearrangement, retro 2+2 cycloaddition, and de novo induction of asymmetry in meso intermediates). The accompanying paper by Janda, Shevlin, and Lerner (*9*) has carried the emerging field of antibody catalysis to new and potentially important terrain. Catalysis by an antibody has been shown to bring into being an otherwise disfavored reaction.

Thus, substrate **1** has been synthesized (see box). In the absence of antibody catalyst (see discussion below), this compound would give rise to the exo-cyclization-derived product **2**. However, in the presence of antibody, the conversion of **1** to **3** is the one observed. As is the case with many innovative advances, the feasibility, in principle, of such an outcome should hardly have been questioned. To the extent that catalysis (antibody or classical) could be specifically directed to the conversion **1** to **3**, the otherwise disfavored reaction may compete with, or even overwhelm, the background (favored) reaction of **1** to **2**. I shall return to the logic used by Janda *et al.* in eliciting the antibody to drive the disfavored reaction (*9*). However, to grasp the advance more fully, it is well to digress and review the intellectual history of the "cyclization trajectory" problem, from the standpoint of organic chemistry. It is to this issue that antibody catalysis was applied by the Scripps workers.

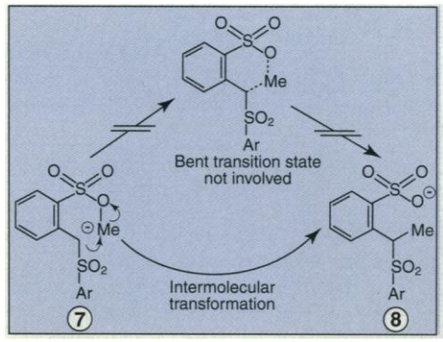
As with many areas, the epistemological starting point for the analysis is not rigorously defined. Certainly the stereochemical concepts of Walden inversion had been part of the standard perception level of organic chemists (*10*) for 50 years (see **4**+**5** → **6**). Also part of the intellectual fabric of the field was the notion that intramolecular processes are much more facile than their intermolecular counterparts, presumably for entropic reasons (*11*).

In 1970, Eschenmoser and co-workers described experiments in which the truisms of preferred intramolecularity and preferred colinearity were pitted against one another (*12*). For the transformation of **7** to **8** to be

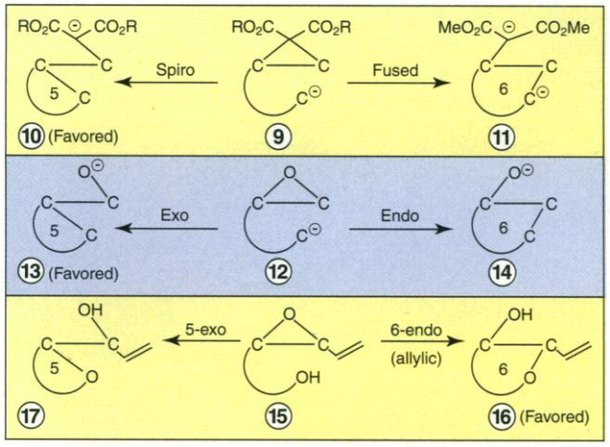


The author is in the Department of Chemistry, Yale University New Haven, Connecticut 06511-8118.

intramolecular (as is seductively suggested by the curved arrows in the box at right), serious departure from the preferred 180° bond angles would be necessary. Describing a classical study, rich in imagination while fully rigorous in experimental and analytical standards (a combination not to be taken lightly), the Helvetica paper (*12*) concluded that the trajectory requirements of approximate colinearity override the classical preference of intramolecularity. The transformation of **7** to **8** which does take place is, in fact, bimolecular.



The next step arose from our laboratory, then in Pittsburgh (*13*). The principles articulated in the Eschenmoser paper (*12*) were used to direct ring formation of unfavorable size, by imposing significant deviations from linearity upon the otherwise favored cases. My co-workers and I have analyzed this problem in terms of "spiro" (**9** to **10**) versus "fused" (**9** to **11**) transition states (*13*).



We naïvely thought our teachings to be particularly straightforward in their implications. Nonetheless, much of the didactic value of our papers was apparently lost on later commentators, presumably because our examples dealt with a rare type of displacement reaction, wherein the leaving group was a carbon atom of an activated cyclopropane substrate. A more widely received statement of the trajectory principle as applied to this kind of ring mutation was articulated by Stork and co-workers (*14*). In these cases, the attacking nucleophile was once again a carbon anion, but the leaving group was the oxygen of an epoxide. The Stork work demonstrated that both the exo (as for spiro) **12** to **13** and endo (as for fused) **12** to **14** modes were possible. However, at roughly equal levels of steric hindrance, the exo mode was preferred even when this leads to an otherwise highly strained cyclobutane.

From these and other experiments there then followed the articulation of the "Baldwin rules" of ring closure (*15*). These correlations went well beyond the sp^3 cases of Eschenmoser (*12*), Danishefsky (*13*), and Stork (*14*) described above. Baldwin attempted, in a heroic and global fashion, to codify a range of cyclization reactions (including those occurring sp^2 and sp centers). Not unlike the case with many promulgations, attempts at increasing inclusion necessitated sometimes awkward recourse to provisos. However, the contributions of the Baldwin school in facilitating the systematics of reasoning vis-à-vis the all important question of cyclization reactions in synthesis can hardly be exaggerated.

From these previous works, there arose a high level of confidence that a hydroxyalkyl epoxide of the type **1** would undergo favored cyclization to **2** in the 5-exo (spiro) rather than to **3** in the 6-endo (fused) sense. Even here it must be emphasized that structural nuances can overcome such principles. Thus, in an elegant demonstration, Nicolaou and co-workers described preferred transformations of the type **15** to **16** (*16*). Hence, allylic substitution can render the 6-endo (fused) mode preferred relative to the 5-exo pathway. Also to be studied is an ingenious series of experiments by Beak and co-workers (*17*) that probe the question of the exo versus endo modes of displacement in substrates that lack the conventional stereochemical markers to assess the sense of attack.

Enter Janda, Shevlin, and Lerner (*9*). The transition analog substrate moiety was incorporated in the hapten (see **18**) to stimulate immune response. Antibodies, thus elicited, were confronted with hydroxyepoxide system **1**. It was hoped that **1** would be directed by the binding capacity of the antibody combining site, toward the transition state favoring the formation of product **3**. Indeed a particular antibody, 26D9, accepted only one enantiomer of **1** giving rise to enantiomerically pure **3**.

It is interesting that the antibody combining site, fashioned by the immune system to bind tertiary amine oxide hapten **18** (see box on previous page), is as selective as it is in guiding substrate **1** in the direction of **19**, a model for the presumed pre-**3** transition state. The charge distributions, not to speak of the relevant atoms bearing the charges in **1**, are clearly different from those of presumed transition state **19** leading to **3**. A decisive role for the catalytic antibody cannot be challenged since, in the absence of protein, formation of expected product **2** is highly favored. Insight as to the specifics of how antibody 26D9 accomplishes this result would be of great interest but must await precise structural analysis of the protein. One can be confident that the story, when fully understood, will be fascinating.

Also, if history be a guide, the finding of antibody catalysis of a "disfavored" reaction will result in growing demands for "encores" which will involve more difficult challenges. Included in such an early wish list might be an exo Diels Alder reaction, an anti-Markovnikov addition to a double bond or a syn bromination. Indeed, can an antibody catalyst be elicited which would impose the condition of intramolecularity on the Eschenmoser transformation, that is **7** to **8**?

The ability to render the disfavored reactions of organic chemistry accessible to the practicing chemist is a potential spin-off of the Janda, Shevlin, Lerner paper (*9*). New successes in this regard would be eagerly received by chemists. Not the least benefit would be the realization of new strategies in transition state analog-hapten design. Such advances would lead to increasingly refined insights into the catalysis of chemical reactions, a most worthy subject.

References and Notes

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