Antibody-Catalyzed Cationic Reactions: Rerouting of Chemical Transformations via Antibody Catalysis

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Introduction

Since the first papers on catalytic antibodies were published, a large number of antibodies have been developed that catalyze a wide range of reactions.[1] However, prior to our work, antibody-catalyzed cationic reactions had not been reported.[1] Carbocations can be difficult to generate and are such highly reactive intermediates that it is not easy to predict or control their reaction pathways.[2] This is especially true for biochemical systems because a carbocation can interact with a protein's peptidic side chain(s) and/or its backbone, thereby nullifying the required activity and alkylating the protein. Nonetheless, nature deals very efficiently with carbocations. A number of enzymes are known to catalyze cationic processes.[3] One of the most remarkable is 2,3-oxidosqualene cyclase.[4] This enzyme catalyzes the formation of a highly sophisticated tetracyclic triterpenoid with a concurrent establishment of seven asymmetric centers.[3a,5]

For more than 30 years, chemists have been trying to simulate cationic cyclization processes.[6] This work has led to the recognition that cationic cyclization reactions can be divided into initiation, propagation, and termination events. Each of these phases must be controlled if one wishes to precisely organize the outcome of the reaction. Typically cationic cyclization reactions are initiated by formation of a putative carbocation either by electrophilic addition to a double bond or by ionization, usually from an sp^3-hybridized carbon. Here protonic and Lewis acids have been the most frequently used electrophiles.[7,8] Where cyclohexane rings are formed, the overwhelming number of cases can be interpreted as forming *via* a chairlike transition state. Furthermore, where ring junctions are formed, most examples are of (*E*)-alkenes cyclizing to form trans-fused rings to give the thermodynamically most stable product. Termination can be achieved by elimination and/or attack by an internal or external nucleophile.[9]

For antibodies to serve as catalysts for the initiation and control of cationic cyclization reactions, they must be capable of catalyzing a reaction in which it is necessary to simultaneously stabilize point charges, overcome entropic barriers, and provide a chiral binding pocket to generate the desired stereoselectivity.[10] In essence the problem reduces to that of generating a carbocation in an environment that stabilizes its formation and controls its subsequent reaction pathway. The following is an Account of our research directed toward antibody-catalyzed cationic reactions.

Cationic Cyclization Reactions: Solutions for Generating Antibody Monocyclization Catalysts

A "Bait and Switch" Approach. Our work was directed toward the simplest model system, a cationic monocyclization reaction. Drawing from the classic system investigated by Johnson,[11] we designed a sequence in which the initiating carbocation is formed by the solvolysis of a sulfonate ester (Figure 1). In this scenario once cyclization takes place, the newly formed carbocation can be captured through either elimination or attack by an internal or external nucleophile. The dimethylphenylsilyl moiety contained within **1** was included to provide mechanistic information about the reaction pathway in that it offered yet another alternative for capture of the carbocation *via* loss of the silane and formation of an olefin, a process which readily occurs in organic solvents.

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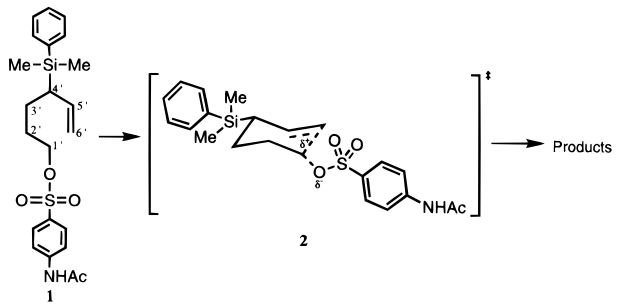
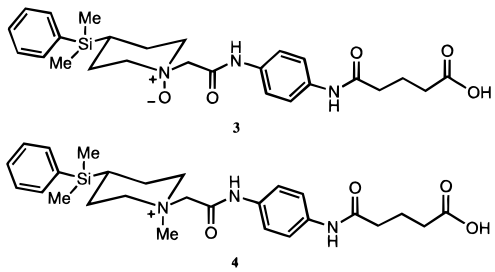


1 2

FIGURE 1. Cationic cyclization reaction and a plausible transition state for the reaction.



3

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FIGURE 2. Haptens used to generate catalytic antibodies for cationic cyclization reactions.

In addition, the outer shell electrons of the silane might be expected to participate in the cyclization by resonance stabilization of the incipient carbocation β to the silicon (β-effect).[12] In the uncatalyzed "Johnson case" a solvolysis−cyclization reaction occurs that is not useful because of poor yield and formation of a complex mixture of products.[11] At acidic pH several zwitterionic and/or cationic species can develop in the transition state of which **2** is representative. However, we believed that antibody catalysis could provide selective control in the mechanism of the solvolysis of **1**, thereby reducing the complexity of the reaction and improving the yield of desired products.

To catalyze the cationic cyclization of **1**, haptens[13] were designed to elicit antibodies that simultaneously facilitate the cleavage of the sulfonate and control the conformation of the substrate in the transition state such that the olefin is properly aligned to participate in the reaction. The cyclic *N*-oxide **3** seemed to be a good candidate hapten to elicit antibodies capable of catalyzing the release of the sulfonate from **1** (Figure 2). Hapten **3** contains an anionic oxygen atom that can elicit a complementary charge in the antibody combining site capable of operating *via* a process that we have termed "bait and switch" catalysis to stabilize the developing negative charge on the departing sulfonate.[14] Such interaction with the leaving group is necessary because our sulfonate ester is not as activated as the nosylate utilized in the original Johnson system. The cationic nitrogen can be expected to induce an anionic functionality in the antibody combining site that should stabilize the developing carbocation so that it is not prohibitively high in energy and hence reduce its inherent reactivity. We envisioned that the reactants would adopt a quasi-chairlike conformation in the transition state with the leaving group in the pseudoequatorial position with the olefinic bond aligned to participate in what is essentially a concerted transformation (Figure 1). The expected chair conformation of the cyclic *N*-oxide hapten **3** should induce antibodies which favor a similar conformation of the reactant in the transition state, thereby facilitating the energetically favorable route for the cyclization process. Proper alignment of the olefin is further assured by conjugation with the equatorial carbon−silicon bond which correctly positions the π orbital for backside attack on the carbon atom. We also synthesized the *N*-methyl quaternary ammonium ion **4** as an alternate hapten for the same reaction. Compared to the *N*-oxide hapten **3** which contains only partial charges, a full positive point charge exists in this hapten.

Both haptens were prepared from 4-bromopyridine hydrochloride and 1,4-phenylenediamine.[15,16] Because the overall shape of the haptens was important to this study, we used nuclear magnetic resonance (NMR) to study the conformation of **3** and **4** in solution. The silicon appendage and the phenylenediamide moiety were unambiguously assigned to the equatorial positions by nuclear Overhauser (NOE) measurements. While the axial orientation of the oxygen atom in **3** might not be considered optimal for induction of a functionality for stabilization of a pseudoequatorial leaving group, the distances between a pseudoequatorial and an axial atom are close enough so as to expect stabilization by any induced complementary functionalities.

The haptens were coupled to the protein carrier keyhole limpet hemocyanin (KLH), and the conjugates were used to immunize 129 G1X$^+$ mice for production of monoclonal antibodies.[17] Twenty-nine monoclonal antibodies were obtained for hapten **3**, while 18 monoclonal antibodies were obtained for hapten **4**. On the basis of a high-performance liquid chromatography (HPLC) assay, four antibodies (4C6, 16B5, 1C9, 6H5) elicited to the *N*-oxide hapten **3** were identified as potential "initiating catalysts" on the basis of their ability to catalyze the cleavage of the sulfonate ester bond in **1**, while one antibody catalyst (87D7) from the quaternary ammonium ion hapten **4** exhibited a similar solvolysis product. The nature of the terminated products was analyzed by gas chromatography (GC). In the case of antibodies elicited to the *N*-oxide hapten **3**, one (4C6) led to the formation of cyclohexene (**5**) (2%) and *trans*-2-(dimethylphenylsilyl)-cyclohexanol (**6**) (98%).[15] Antibodies 16B5, 1C9, and 6H5 did not yield identifiable products. The catalytic antibody

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(13) Hapten is defined here as a small molecule that is not immunogenic by itself; however, when this molecule is conjugated to a carrier protein, it has the ability to stimulate the formation of antibodies highly specific and complementary to its structure. For a general description of basic immunology and more specifically antibody formation, see: Eisen, H. N. *General Immunology*; J. B. Lippincott Co.: Philadelphia, 1990; p 259.

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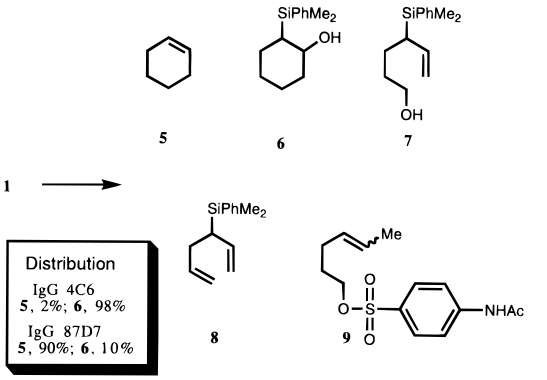

FIGURE 3. Product distribution from the 4C6 and 87D7 antibody-catalyzed reactions.

raised against the quaternary ammonium hapten **4**, 87D7, led to the formation of cyclohexene (**5**) (90%) and *trans*-2-(dimethylphenylsilyl)cyclohexanol (**6**) (10%) (Figure 3).[16] In terms of kinetic parameters both 4C6 and 87D7 can be considered good catalysts for a cationic monocyclization reaction (for 4C6, $k_{cat} = 0.02$ min^{-1}, $K_m = 230\ \mu$M; for 87D7, $k_{cat} = 0.02$ min^{-1}, $K_m = 25\ \mu$M).[18] More striking is that among all possible products (**5**, **6**, **7**, **8**, **9**, Figure 3) as inferred from W. S. Johnson's work, only the cyclized products **5** and **6** were formed. The mass balance of the combined cyclized products (**5** + **6**) also matches the production of 4-acetamidobenzenesulfonic acid in both cases. Both reactions are competitively inhibited by the corresponding haptens, indicating that the reactions occur within the antibody combining site (K_i for 4C6 is 1.0 μM, K_i for 87D7 is 1.4 μM). Quite remarkable is that substrate **1** is extremely stable under our assay conditions (RT, pH 7.0) as no uncatalyzed solvolysis reaction could be observed even after one month of incubation, thus precluding an accurate determination of k_{uncat}.

The overall structural difference among haptens **3** and **4** is slight, yet an almost complete reversal of product ratios between these two catalytic antibodies was observed. For antibody 87D7, generated from hapten **4**, the olefin which results from the elimination of the dimethylphenylsilyl appendage is the dominant product. Stereoelectronic analysis indicates that the silyl group has to adapt to a pseudoaxial position before elimination can occur. It is tempting to hypothesize that the differences in charge between these two haptens are the governing factor for the product differences seen between these two catalysts. We would suggest that much of the binding energy in the case of IgG 87D7 is directed toward charge stabilization. This allows a more relaxed recognition of the silyl appendage or an overall more flexible binding pocket, thereby allowing time for the silyl appendage to rotate to the required position for elimination.[19]

A Transition-State Approach. In haptens **3** and **4**, a formal positive charge is centered on the nitrogen atom, while the actual predicted transition state for a cationic monocyclization reaction is one in which charge is localized along three contiguous carbon atoms (Figure 1). In order to better mimic these stereoelectronics, an amidinium-based hapten (**10**) was synthesized (Figure 4).[20] As shown, hapten **10** possesses a pseudo-half-chair conformation wherein the positive charge is delocalized over three atoms. Although **10** has a greater overall stereoelectronic fidelity to the transition state, its overall geometry, as defined by the amidinium moiety, does not precisely match the starting conformation outlined in the Stork–Eschenmoser hypothesis because of the positions where the two methyl substituents are located.[21] Yet, using such a planar charge approach was thought to alleviate any chance of product inhibition.

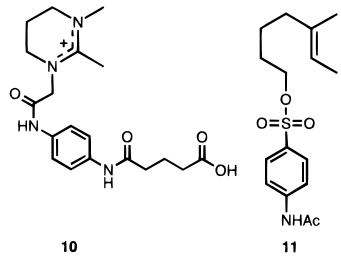

FIGURE 4. Hapten 10 and substrate 11 used in the assay of antibodies elicited to 10.

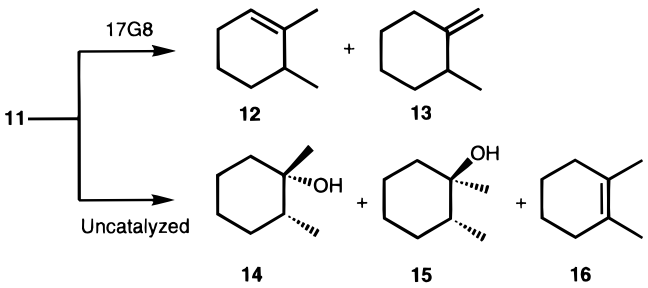

FIGURE 5. Antibody 17G8-catalyzed monoterpenoid formation and the uncatalyzed reaction.

The hydrocarbon unit of **11** mirrors the first two isoprene portions of 2,3-oxidosqualene except that the two methyl substituents at the site of the initial cationic center were omitted to exclude potential elimination reactions. Hapten **10** elicited twenty-four hybridomas, three of which were catalysts (1F8, 12E8, 17G8), based on the release of sulfonic acid. The most active catalyst IgG 17G8 ($k_{cat} = 0.025$ min^{-1}, $K_m = 35\ \mu$M, $k_{uncat} = 3.6 \times 10^{-4}$ min^{-1}) was studied and found to synthesize the carbocyclic compounds **12** and **13** (Figure 5). In the uncatalyzed reaction, two diastereomeric tertiary alcohols (**14**, **15**) and a small amount of 1,2-dimethylcyclohexene (**16**) are formed (Figure 5). On the basis of the product distribution between the catalyzed and the uncatalyzed reaction, it is clear that IgG 17G8 excludes water from the antibody's combining site as well as reroutes the naturally occurring cationic cyclization reaction. Finally, of further note, products **12** and **13** match the core structures of α- and γ-irones, respectively.[22] Irones are the natural terpene constituents of violet-derived perfumes (Figure 6). Depending upon hapten design, in the future it may be possible to utilize

(18) Biphasic solvent conditions: 83% pentane/2% chloroform/15% bicine buffer, 50 mM, pH 7.0. Ashley, J. A.; Janda, K. D. *J. Org. Chem.* **1992**, *57*, 6691.

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(22) Compounds **12**, **13**, and **16** are analogous in their double bond regiochemistry to α-, γ-, and β-irones, respectively. See: Jaenicke, L.; Marner, F.-J. *Pure Appl. Chem.* **1990**, *62*, 1365.

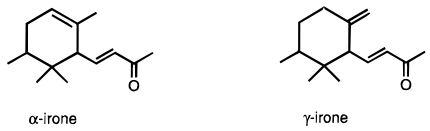

FIGURE 6. Two constituents making up the principle fragrance of violets.

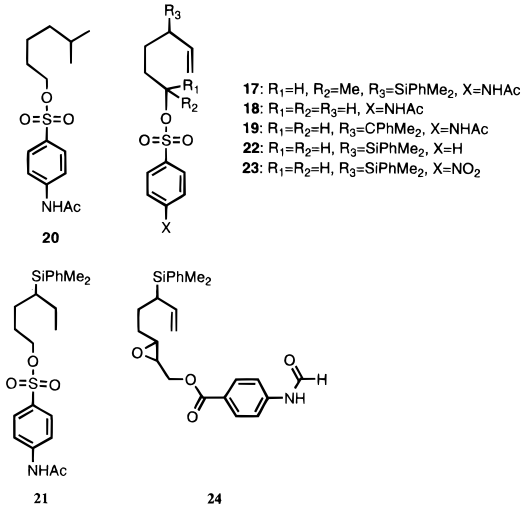

FIGURE 7. Additional substrates to probe the mechanism of the IgG 4C6- and 87D7-catalyzed monocyclization reactions.

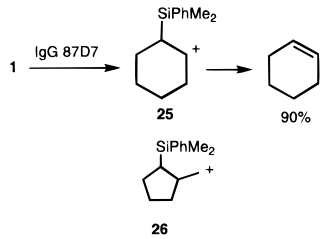

FIGURE 8. Potential reactive intermediates generated from the IgG 87D7-catalyzed reaction.

an antibody to selectively synthesize one of these fragrance principles of violets.

Mechanistic Analysis of Antibody-Catalyzed Monocyclization Reactions

In addition to **1**, other substrates (**17**−**24**) were synthesized to explore the mechanistic aspects of the IgG 4C6- and 87D7-catalyzed reactions (Figure 7). The most interesting comparison using IgG 4C6 was between compounds **1** and **21** which differ only by the presence of an olefin. Only **1** is a substrate for the reaction, thereby providing evidence that the olefin assists in the departure of the sulfonate ester *via* anchimeric assistance and the entire process proceeds most likely by a concerted π route. The failure of the silane to depart also provides mechanistic information about the antibody-catalyzed process. Termination of a fully enforced concerted process with the silane departing from the equatorial position would require formation of the highly unfavorable *trans*-cyclohexene. Thus, we believe that a concerted π route leads to the formation of a discrete carbocation species (**25**) (Figure 8) which terminates by the addition of water or by loss of the silyl group.

Sulfonate **17**, the closest homolog of **1**, was found to be a substrate of 4C6 ($k_{cat} = 0.3$ min^{-1}, $K_m = 1.8$ mM, $k_{cat}/k_{uncat} = 12000$, for sulfonate release), but clean cyclization

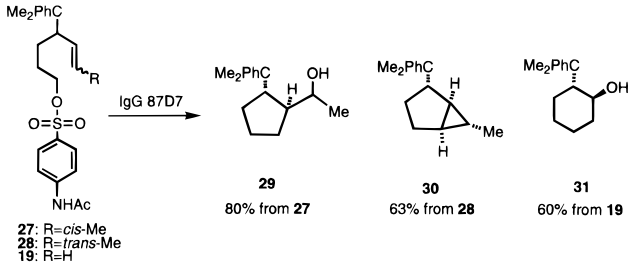

FIGURE 9. Product distribution seen with IgG 87D7 and substrates 19, 27, and 28.

was not observed. Instead, the 4C6-catalyzed reaction of **17** yielded a complex mixture of products. The different reaction outcome for substrates **1** and **17** may simply reflect the variances in reaction pathways open to the reactants. In the catalytic transformation of **1** obligatory anchimeric assistance by the olefin leads to a restriction in product complexity. Such constraints do not pertain to **17** where a more stable secondary carbocation occurs on the reaction pathway. It is well known for many reactions that when the possibility of alternative stabilization of the carbocation exists, the reaction may proceed by a route that no longer involves anchimeric assistance.[23]

Compound **19** which differs from **1** only by substitution of a carbon for a silicon atom not only is a poor substrate for 4C6 ($k_{cat} = 7.8 \times 10^{-4}$ min^{-1}, $K_m = 330$ μM, for sulfonate release) but also yielded a complex mixture of products.[15] This suggests that the silicon atom plays an important role in this antibody-catalyzed reaction probably by activation of the olefin or participation in the overall stabilization of the cationic process or both. However, interestingly, compound **19** is an excellent substrate for 87D7, yielding cyclohexanol **31** in good yield (Figure 9).[24]

Compounds **18** and **20** were ineffective as substrates for antibody 4C6, suggesting that the dimethylphenyl functionality plays an important role in the antibody−substrate recognition. The acetamido sulfonate group in substrate **1** appears to be important also for antibody recognition as the closely related sulfonates **22** and **23** failed to yield cyclized products with either antibody. Thus, the effectiveness of our catalyst appears to be governed by several factors including stabilization of point charges, provision of sufficient binding to overcome entropic barriers, and the more complicated stereoelectronic effects which can be provided by precise control of the conformation of the reactants. Finally, the epoxide-containing compound **24** was not a substrate for the antibody 4C6.[15] Presumably, a lack of similarity with hapten **3** precludes compound **24** from being a substrate for antibody catalysis.

An Unexpected Reaction Product Derived from the Antibody-Catalyzed Reactions: Cationic Cyclopropanation

The antibody-catalyzed reactions described above undoubtedly go through a highly reactive carbocation intermediate whose fate is largely controlled by the antibody

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catalyst. But, **1** also has intrinsic chemical features that play a role in guiding the reaction to produce the more stable secondary carbocation **25** rather than the primary carbocation **26** (Figure 8). We therefore envisioned that by making subtle structural modifications to the substrate, we could intervene in the reaction to direct the formation of a variety of other products. Thus, the silicon atom of the phenyldimethylsilyl group could be replaced with a carbon atom so that a potential β-effect would no longer influence the reaction pathway. Also, a methyl functionality could be added to the terminal olefin appendage of **1**, thereby eliminating the chemical advantage of a reaction route involving a secondary rather than a primary carbocation.

On the basis of the above assumptions, the *cis*-olefin **27** was synthesized.[24] When **27** was incubated with IgG 87D7, clean formation of a single diastereomeric exocyclic alcohol (**29**) was observed (Figure 9, $k_{cat} = 0.013$ min^{-1}, $K_m = 58$ μM). The structure of **29** was confirmed by comparing both the proton NMR spectrum and the GC trace of the sample obtained from the antibody-catalyzed reaction with a racemic sample synthesized independently.[24] However, the most interesting result was obtained when the *trans*-olefin **28** was used as a substrate in that formation of **30** in 63% yield was observed (Figure 9, $k_{cat} = 0.021$ min^{-1}, $K_m = 102$ μM). Importantly, the rate acceleration achieved by this antibody catalyst is within an order of magnitude of those of natural enzymes that catalyze similar cationic processes.[25] The most salient feature of **30** is the strained cyclopropane ring found in the bicyclo[3.1.0]hexane unit. As with **29**, we confirmed, **30**'s structure by the direct comparison of both the proton NMR spectrum and the GC trace of the sample obtained from the antibody-catalyzed reaction to an authentic synthetic sample. To prove that the cyclopropanation reaction was dependent on antibody catalysis, substrate **28** alone was subjected to Johnson-like conditions (formic acid/sodium formate, 80 °C, 2 h)[4] and biphasic conditions comparable to those used for antibody catalysis (hexane/bis-tris, pH 2, 80 °C, 36 h or hexane/DMSO/bis-tris, pH 2, 80 °C, 12 h). The extreme pH and temperatures employed were necessary as in the absence of the antibody no solvolysis was observed. Yet, even under these harsh conditions, no cyclopropane product could be detected, thereby demonstrating that antibody catalysis is essential for its formation.

A unified reaction pathway invoking protonated cyclopropane **32** (Figure 10) can be assigned which rationalizes the reaction products observed in the reaction shown in Figure 9.[26] Thus, for **19**, addition of water to the α-carbon of **32** leads to the formation of cyclohexanol **31**. In the case of **27** and **28** which contain an electron-donating methyl group, product **29** or **30** is formed by addition of water to the β-carbon of **32** of loss of a proton from **32**, respectively. The product partitioning seen can thus be ascribed to the antibody exerting control over the stereoelectronic properties of the carbocation intermediate. In

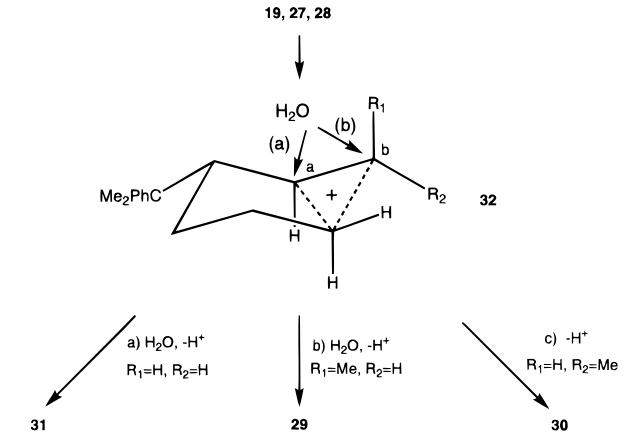

FIGURE 10. Protonated cyclopropane 32 as a reactive intermediate for antibody 87D7-catalyzed reactions. Reprinted with permission from ref 24. Copyright 1996 Macmillan Magazines Limited.

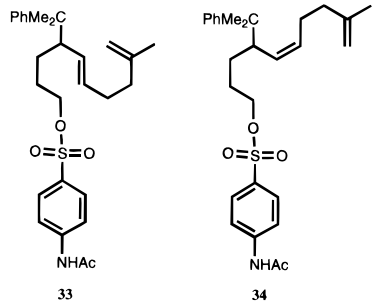

FIGURE 11. Substrates examined for tandem cationic cyclization reactions.

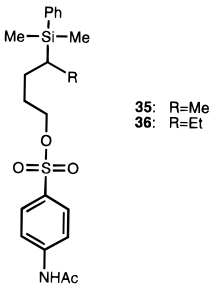

FIGURE 12. Substrates investigated with IgG 16B5 for nucleophilic substitution reactions.

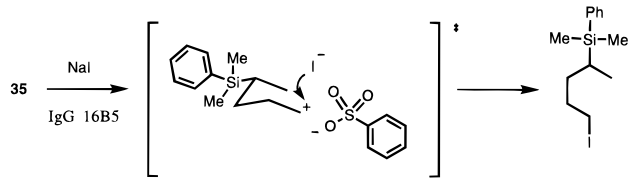

FIGURE 13. Our suggested reaction mechanism for the antibody 16B5-catalyzed nucleophilic substitution reaction.

essence we see that subtle structural changes in the substrate can allow the antibody to redirect the chemical transformation such that alternative reaction routes can be traversed.

The antibody-catalyzed cationic cyclopropanation reaction is important for two reasons. First, the cationic cyclopropanation reaction is usually a highly disfavored process in chemical systems.[27] To the best of our knowledge, cyclopropane formation has not been reported in the numerous attempts to mimic 2,3-oxidosqualene cyclase using chemical systems. Indeed our failure to see

(25) Lewinsohn, E.; Gijzen, M.; Croteau, R. *Arch. Biochem. Biophys.* **1992**, *293*, 167.
(26) (a) Olah, G. A. *Angew. Chem., Int. Ed. Engl.* **1995**, *34*, 1393. (b) Brown, H. C. *Pure Appl. Chem.* **1982**, *54*, 1783−1796.

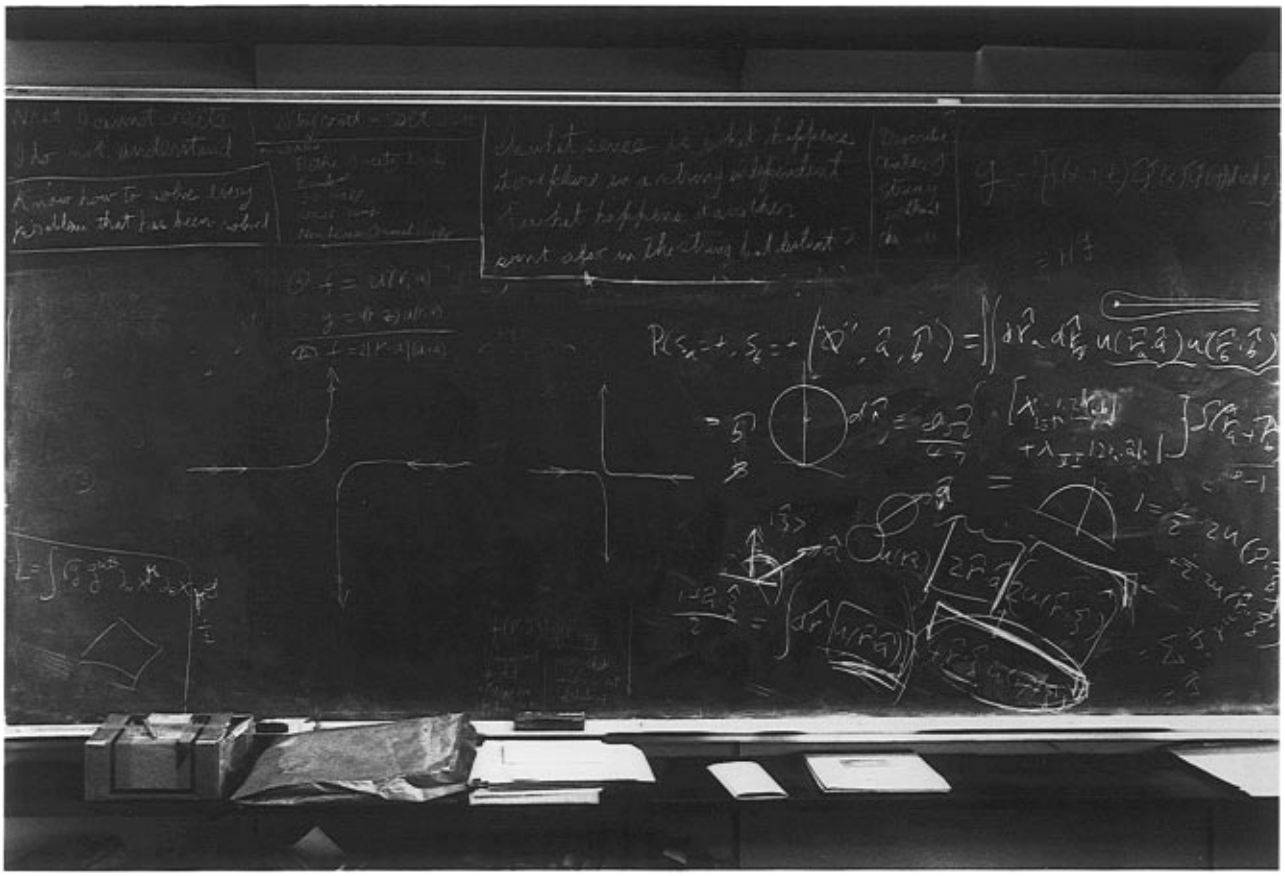

FIGURE 14. Photograph of Professor Feynman's blackboard taken in 1988 at the California Institute of Technology (see upper left-hand corner for quote). Reprinted with permission (see ref 35).

the cyclopropane product in solvolysis experiments with **28** without an antibody catalyst again illustrates the difficulty in obtaining this product.

Tandem Cationic Cyclization Reactions

Compounds **33** and **34** were synthesized to explore the possibility of an antibody bicyclization reaction which would be a closer mimic to 2,3-oxidosqualene cyclase (Figure 11). Preliminary results indicate that 87D7 was active in catalyzing the sulfonate-cleavage in both substrates while 4C6 showed no detectable activity. The nature of the cyclized products has yet to be determined.[28] Other efforts in using bicyclic haptens to effect tandem cyclization are also under way in our laboratories.[29]

Beyond Cationic Cyclization Reactions: Nucleophilic Substitution Reactions through Contact Ion Pairs

Nucleophilic substitutions are one of the most thoroughly studied and best understood reactions in organic chemistry. Both S_N1 and S_N2 mechanisms are operative for reactions occurring at aliphatic carbon atoms. While an S_N2 mechanism is usually displayed when substitution takes place on a primary carbon, the S_N1 mechanism predominates in reactions involving a tertiary carbon center. In contrast to chemical reactions, the mechanisms by which nucleophilic substitution occurs in biological systems are poorly understood. Antibody catalysis allows us to look at the problem because one can utilize highly specific binding energy to isolate reaction routes from an otherwise complex spectrum.[30]

As we have described (*vide supra*), several antibodies raised against the *N*-oxide hapten **3** catalyzed the cleavage of the sulfonate group in substrate **1**. One antibody, 16B5, was interesting because while it was not a cyclization catalyst, it was alkylated during the reaction, suggesting the formation of a reactive substrate carbocation in the antibody combining site. In contrast to its alkylation with olefinic substrates, this antibody catalyzed nucleophilic substitution of the 4-acetamidobenzenesulfonate group in substrate **35** (Figure 12) with sodium iodide.[31] The catalyst exhibited multiple turnovers, no product inhibition ($k_{cat} = 0.028$ min^{-1}, $k_{cat}/k_{uncat} = 580$ M, K_m for NaI at saturating substrate **35** concentration = 150 mM, K_m for substrate **35** at 150 mM NaI concentration = 130 μM).[31] The catalyzed reaction is competitively inhibited by hapten **3** with a K_i of approximately 10 μM.

From an analysis of stereochemical outcome of this reaction, its dependence on the nature of the nucleophile, and secondary isotope effects, we concluded that this

(27) (a) Bayless, J. H.; Friedman, L. *J. Am. Chem. Soc.* **1967**, *89*, 147. (b) Silver, M. S. *J. Am. Chem. Soc.* **1960**, *82*, 2971. (c) Skell, P. S.; Starer, I. *J. Am. Chem. Soc.* **1960**, *82*, 2971. (d) Beaton, J. M.; Easton, J. D.; Macarthur, M. M.; Spring, F. S.; Stevenson, R. *J. Chem. Soc.* **1955**, *Part 4*, 3992.

(28) Li, T.; Lerner, R. A.; Janda, K. D. Unpublished results.

(29) The synthesis of a tricyclic hapten intended for an antibody-catalyzed cationic tandem cyclization reaction has been reported. However, no antibody catalysis was observed. Bell, I. M.; Abell, C.; Leeper, F. J. *J. Chem. Soc., Perkin Trans 1* **1994**, 1997.

(30) Janda, K. D. *Biochem. Soc. Trans.* **1993**, *21*, 1090.

(31) Li, T.; Janda, K. D.; Hilton, S.; Lerner, R. A. *J. Am. Chem. Soc.* **1995**, *117*, 2367.

reaction proceeds via a contact ion pair mechanism.[31] In this model we suggest that 16B5 catalyzes the formation of a contact ion pair within substrate **35** as outlined in Figure 13. In the absence of a properly bound nucleophile, a nonproductive equilibrium exists between the contact ion and **35**. However, when a properly oriented nucleophile is allowed to attack the contact ion from the backside, nucleophilic substitution products are observed. This mechanism is further supported by the fact that compound **36** failed as a substrate for this reaction. In accordance with our hapten design, binding of substrate **36** (Figure 12) in a chair conformation in the antibody cleft would block backside attack with the extra methyl group and thus preclude product formation. The key feat of the catalytic antibody in this reaction sequence is to neutralize the charge while not becoming alkylated. In the case of our catalyst this appears to be accomplished by the combined use of countercharges and the proper positioning of the iodide atom.

(32) (a) Jencks, W. P. *Catalysis in Chemistry and Enzymology*; McGraw-Hill: New York, 1969; p 288. (b) Wooley, D. W. *A Study of Antimetabolites*; John-Wiley & Sons, Inc.: London, 1952; p 87.
(33) (a) Tramontano, A.; Janda, K. D.; Lerner, R. A. *Science* **1986**, *234*, 1566. (b) Pollack, S. J.; Jacobs, J. W.; Schultz, P. G. *Science 1986, 234*, 1570.
(34) Hollfelder, F.; Kirby, A. J.; Tawfik, D. S. *Nature* **1996**, *383*, 60.
(35) Courtesy of the Archives of the California Institute of Technology.

Concluding Remarks

Catalytic antibodies have had a long gestation period from the initial conceptions[32] to the first demonstration.[33] The work reported here demonstrates that catalytic antibodies can accomplish truly exciting deeds. Perhaps most importantly this work illustrates the importance of designing binding pockets as opposed to simply using preexisting scaffolds.[34] The power of antibody catalysts to control chemical transformations stems from the merger of an understanding of a reaction mechanism with the ability of the immune system to yield binding proteins programmed to interact in highly specific ways. In our attempts to understand and make catalysts we suscribe to the notion of Feynman as written on his blackboard (Figure 14): "what I cannot create I do not understand".[35] Ultimately we hope to understand catalysis by making new catalysts!

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