Feb 28 2014

Richard Lerner

Kelvin Chan

Lerner’s career and his work on catalytic antibodies

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Biographical Info:
•Northwestern BSc, 1956–1959
•Stanford MD, 1959–1964
•Stanford hospital Internship, 1964–1965
•Scripps Clinic postdoc, 1965–1968

Key Professional Appointments:
•1968 Associate, Wistar Institute, Pennsylvania
•1970 Associate, Scripps Clinic RI
•1982 Chairman, Scripps Clinic RI
•1987 Director, Scripps Clinic RI
•1988 First President, TSRI

Selected Awards:
•1991 Arthur C. Cope Scholar
•1994/95 Wolf Prize
•1996 CA Scientist of the Year
•2002 Scientist of the Year, ARCS
•2002 UC President's Medal
•2012 Prince of Asturis
•As of 2007, 67 patents, 403 papers

Board Seats:
5AM Ventures, Acro, Bay City Capital, Dyadic, Intra-Cellular Therapies, Kraft Foods, OPKO Health, Optimer, Sequenom, Sorrento Therapeutics

Illustrious career spanning >40 years

Chronology:
1. **Plasma membrane proteins, 1971–1981**
2. **Antibodies and specificity; synthetic peptides, 1981–**
3. **Catalytic antibodies, 1986–**
4. **Combinatorial antibody libraries, 1991–**
5. **Cis-9, 10-octadecenamide, 1994–1997**
6. **Ozone in human disease, 2002–2006**

Collaborators at Scripps:
Peter Schultz, Kim Janda, Carlos Barbas, Dale Boger, Ben Cravatt, Ian Wilson, Frank Chisari, Peter Wright, and many more.

Wolf Prize: "for converting antibodies into enzymes, thus permitting the catalysis of chemical reactions considered impossible to achieve by classical chemical procedures" (joint award with Schultz)

Lerner was the pioneer of catalytic antibodies, and over the next two decades, he developed a strategy to accelerate and catalyze chemical reactions for which traditional methods are not efficient.

This presentation only covers until 2007, and focuses on catalytic antibodies.

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Catalytic Antibodies

Study of immunochemistry – the branch of biochemistry concerned with the immune response and immune system

What are antibodies?

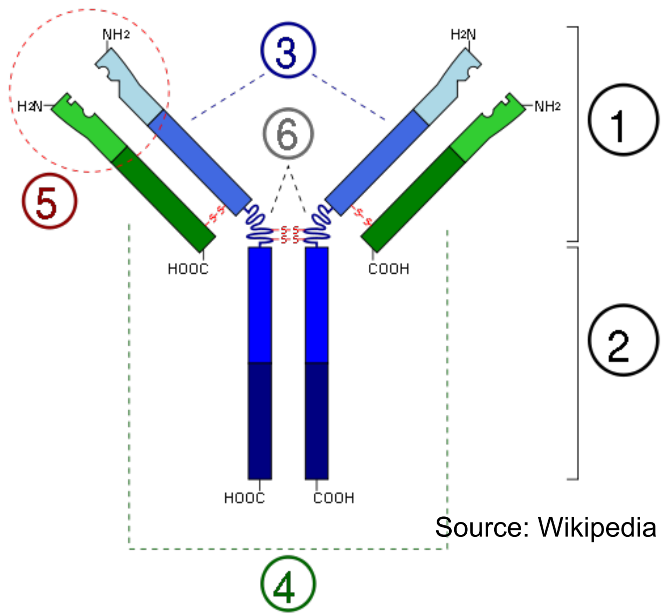
•Also known as immunoglobins (Igs)
•Protein used by the immune system to identify and neutralize foreign objects
•They do so by recognizing the **antigen**, which is the unique part of the foreign target
•Made by B-lymphocytes

Advantages of antibodies as catalysts, Science **1991**, 252, 659

It is possible to tap the cells of the immune system to produce antibodies that bind to any molecule of interest, with high affinity and selectivity.

1. Enormous molecular diversity of immune system makes antibodies highly specific
2. Structural framework is the same – makes antibodies easy to purify, and easy to conduct structural studies, biochemical engineering, bacterial expression
3. Can function in organic solvents by solubilization in reverse micelles
4. Can be immobilized – and still retains activity and specificity in organic solvents

First examples of antibody catalyzed chemical reaction were by Richard Lerner and Peter Schultz in a back-to-back publication in Science **1986**, 234, 1566



Source: Wikipedia

Regions of an antibody:

1. Fab (fragment, antigen binding)
2. Fc (fragment, crystallizable)
3. Heavy chain
4. Light chain
5. Antigen binding site
6. Hinge regions

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Generating catalytic antibodies

Principles

•Basic principle of enzyme catalysis: strong binding interactions are required to reduce the energy barriers along the chemical reaction pathway Typically lowers energy of intermediate.

•Transition-state stabilization, proximity effects, general acid and base catalysis, electrophilic and nucleophilic catalysis, strain etc. are used.

Hapten

•Definition: Small molecule that can elicit an immune response only when attached to a large carrier (e.g., protein)

•Use of hapten elicits desired antibodies

•Hapten thereafter behaves as inhibitor in the catalytic system

Process

"Bait and switch" – the hapten serves as "bait" for attracting catalytic functions in the induction of the antibody; it is then "switched" for the substrate. This can be used even for cofactor approaches.

Strategies

1. Using antibodies to stabilize negatively and positively charge transition states (Pauling)
- TS mimic – both stereo (geometry) and electronically (developing charge)
2. Using antibodies as entropic traps
3. Generating antibodies with catalytic groups and cofactors in their combining sites
- Chemical cofactor is non-covalently bound by antibody *together with substrate* in order to provide chemical reactivity in antibody binding pocket.

Science **1991**, 252, 659

ID and purification

Sometimes the antibodies are identified by their specific reaction with a potential ester substrate that releases a fluorescent product. More commonly now by ELISA. Inject into mice to produce ascites fluid, and isolated/purified. Monoclonal only (made by same immune cells). Verify antibody by PAGE.

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Transformations catalyzed by catalytic antibodies

Many classes of reactions have been demonstrated by Lerner:

1. Hydrolytic reactions (16 publications)

a) Ester hydrolysis
b) Amide hydrolysis
c) Enol ester hydrolysis
d) Enol ether hydrolysis (including glycosidic bond hydrolysis)
e) Phosphate triester hydrolysis

2. Carbon-carbon and carbon-heteroatom bond forming reactions (29 publications)

a) C–N Amide formation
b) C–O Ring closure
c) C–O Epoxidation
d) C–C Diels Alder reaction
e) C–C Cationic cyclizations
f) C–C Rearrangement reactions
g) C–C Aldol reactions (and retro-aldol)
h) C–C Robinson annulation reactions

3. Others (3 papers)

a) Syn elimination from acyclic
b) Oxidation of water

*papers referenced do not include mechanistic studies

Selected reviews: Science **1991**, 252, 659; ACR **1993**, 26, 391; Science **1995**, 269, 1835; ACR **1997**, 30, 115; ACIE **2002**, 41, 4427

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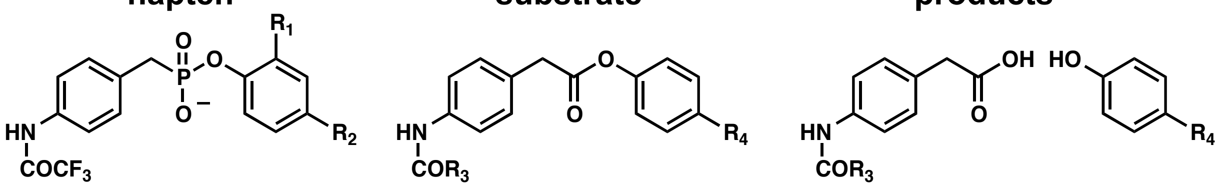
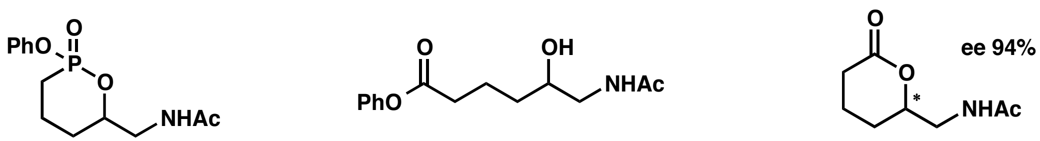
1. Hydrolytic reactions

1a) Ester hydrolysis

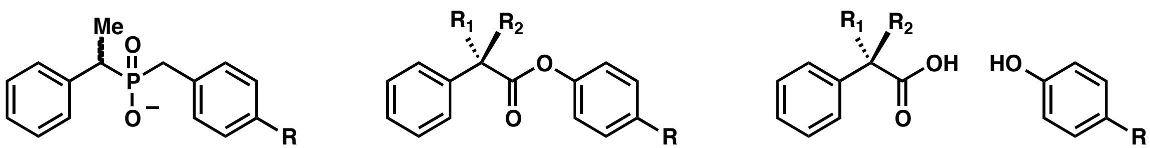
hapten **substrate** **products**



PNAS **1986**, 83, 6736
Science **1986**, 234, 1566
JACS **1988**, 110, 2282



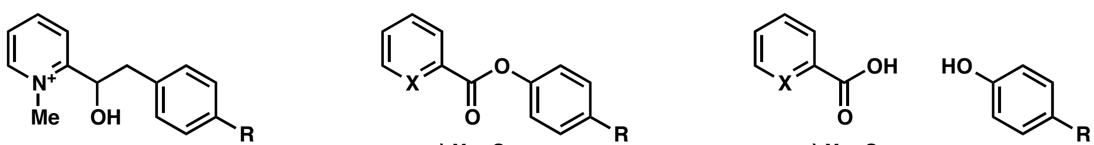
Science **1987**, 237, 1041



a) R_1 = Me, R_2 = H
b) R_1 = H, R_2 = Me

a) R_1 = Me, R_2 = H
b) R_1 = H, R_2 = Me

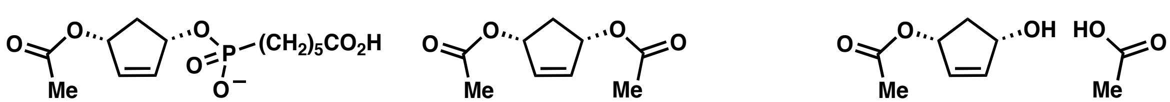
Science **1989**, 244, 437
*Kinetic resolution



a) X = C
b) X = N

a) X = C
b) X = N

JACS **1990**, 112, 1274 (TS)
JACS **1993**, 115, 4906 (cofactor)



JACS **1991**, 113, 7763
*Meso substrate, >98% ee
(background 14%)

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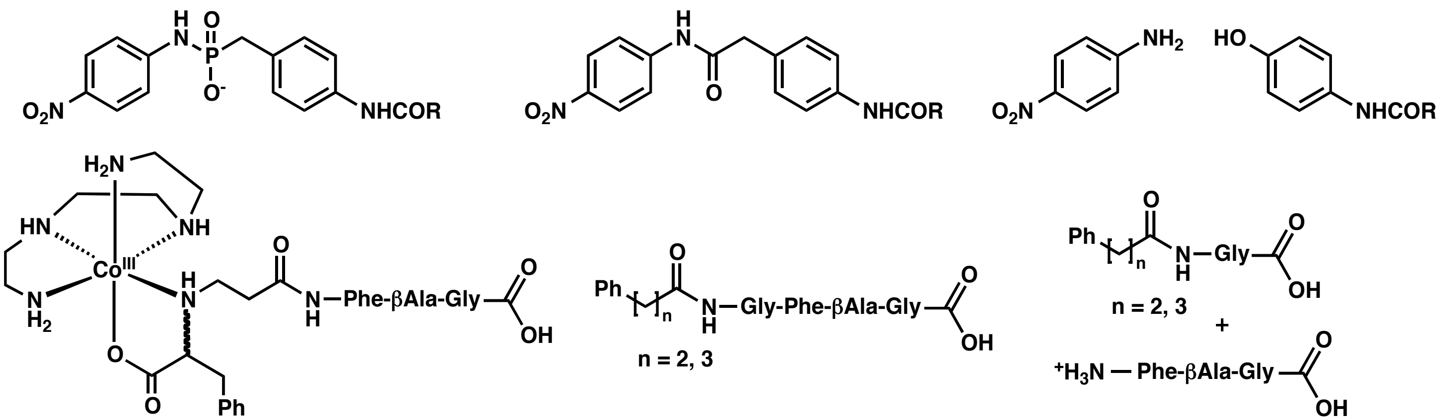
1. Hydrolytic reactions

1b) Amide hydrolysis – kinetically most difficult hydrolysis reaction

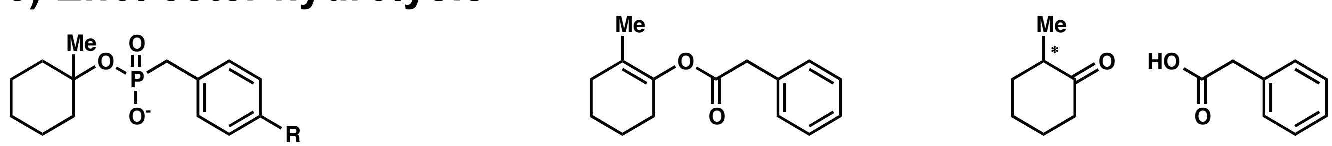
hapten **substrate** **products**



Science **1988**, 241, 1188

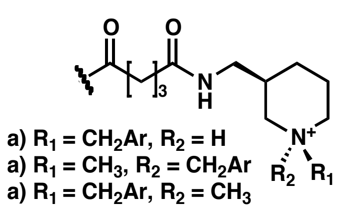
Science **1989**, 243, 1184
*cofactor instead of TS mimic

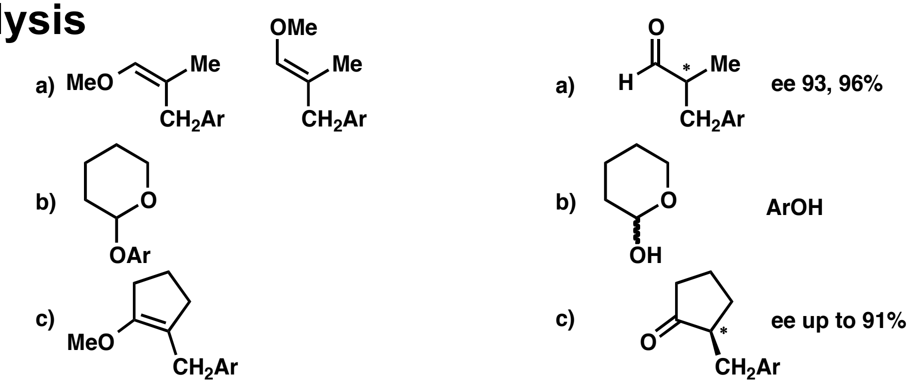
1c) Enol ester hydrolysis



JACS **1991**, 113, 8528
*42% ee, first example of enantiofacial protonation

1d) Enol ether hydrolysis





ACIE **1991**, 30, 1711
JACS **1992**, 114, 2257
JACS **1993**, 115, 3909
ACIE **1994**, 33, 475

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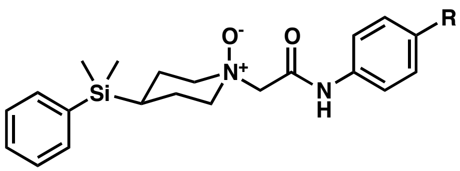
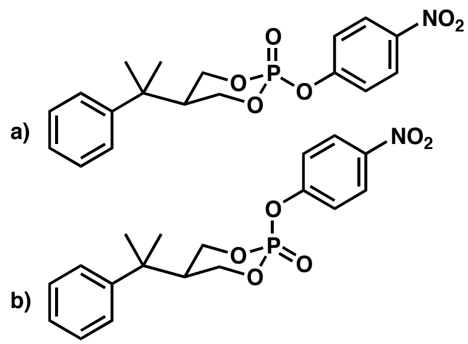
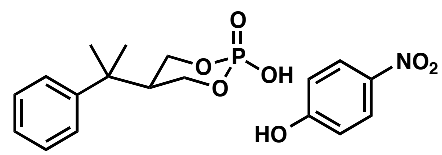
1. Hydrolytic reactions

1e) Phosphate triester hydrolysis

hapten **substrate** **products**



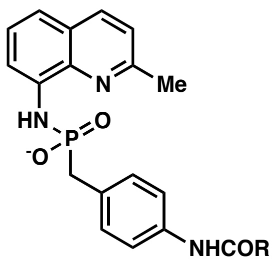


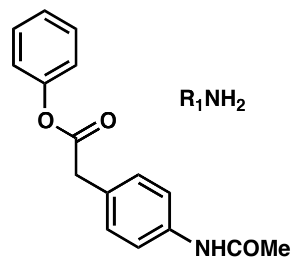


ACIE **1995**, 34, 2275

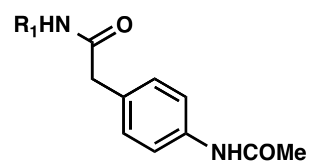
2. C–C and C–X bond forming reactions

2a) C–N Amide formation

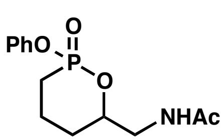


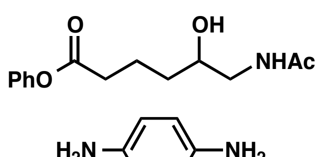


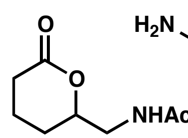
R_1NH_2



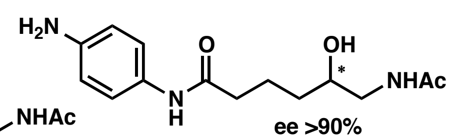
JACS **1988**, 110, 4835







intermediate



PNAS **1988**, 85, 5355

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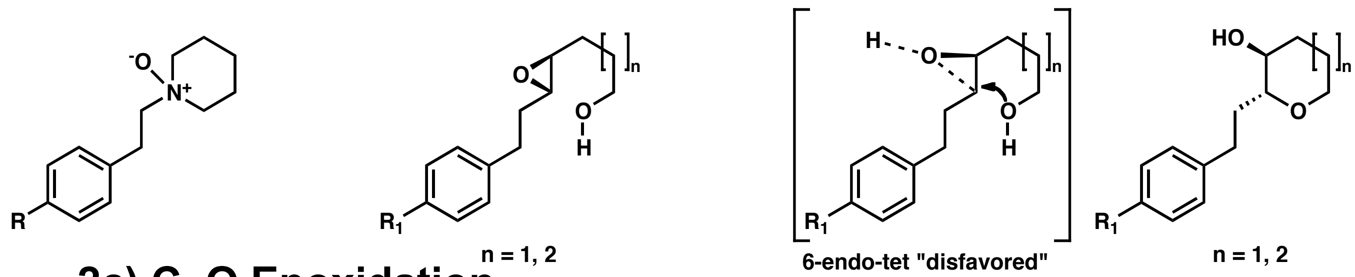
2. C–C and C–X bond forming reactions

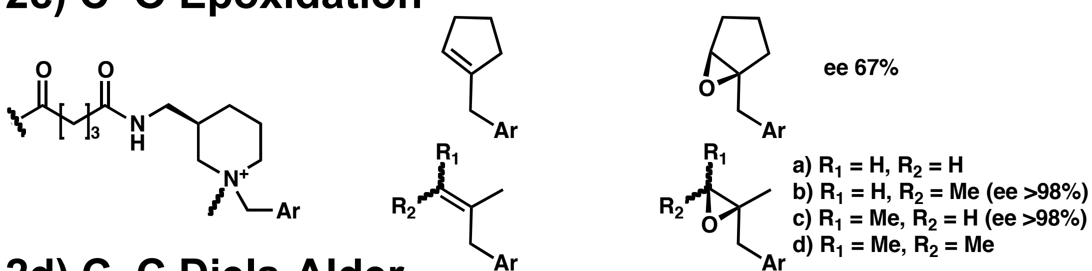
2b) C–O Ring closure (violation of Baldwin's rules)

hapten substrate products



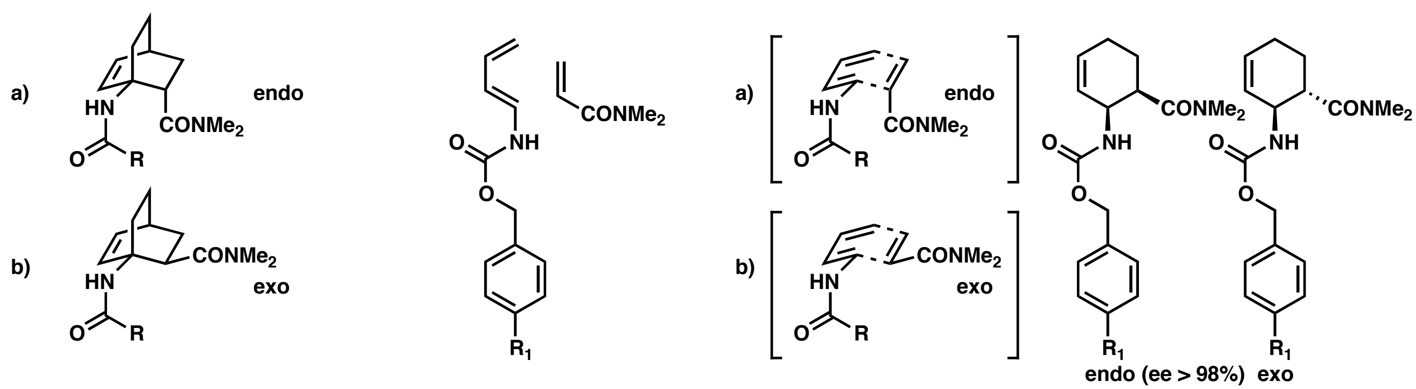
Science **1993**, 259, 490
JACS **1995**, 117 2659

2c) C–O Epoxidation



JACS **1994**, 116, 803
First oxidation reaction at carbon

2d) C–C Diels-Alder



Science **1993**, 262, 204
TS and entropic trap

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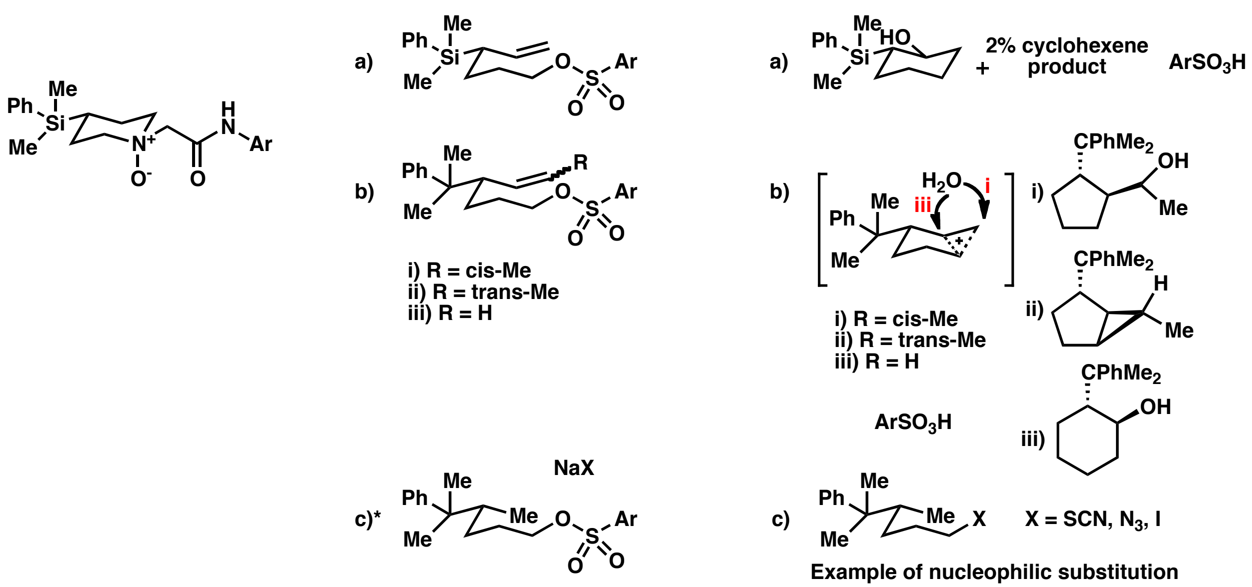
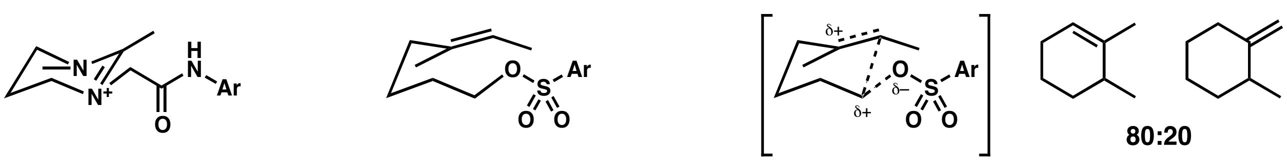
2. C–C and C–X bond forming reactions

2e) C–C Cationic cyclization

hapten | substrate | products



Science **1994**, 264, 1289
Nature **1996**, 379, 326
JACS **1995**, 117, 2367



JACS **1996**, 118, 11654

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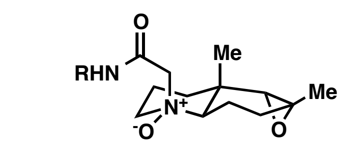
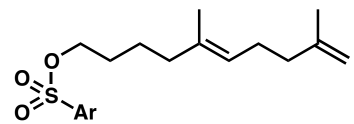
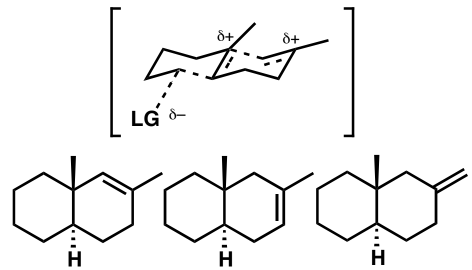
2. C–C and C–X bond forming reactions

2e) C–C Cationic cyclization

hapten | **substrate** | **products**

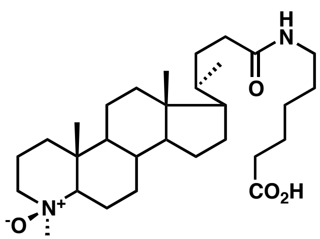


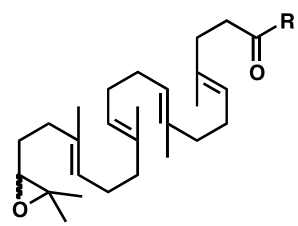




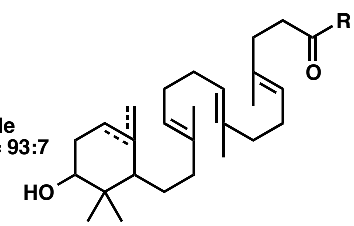
2:3:1

JACS **1997**, 119, 5993
ACIE **1999**, 38, 1743



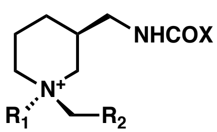


a) R = OH
b) R = NHMe
endo/exo = 93:7

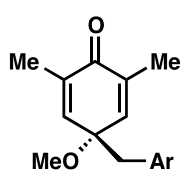


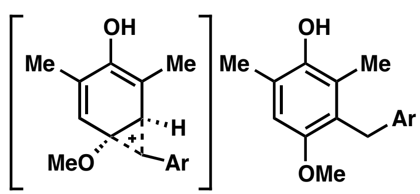
JACS **2000**, 122, 40

2f) C–C Rearrangement reactions



a) R_1 = H, R_2 = Ar
a) R_1 = CH_2Ar, R_2 = H



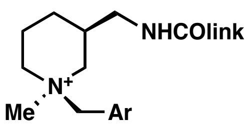


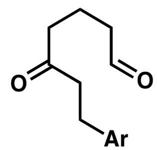
ACIE **1994**, 33, 1607

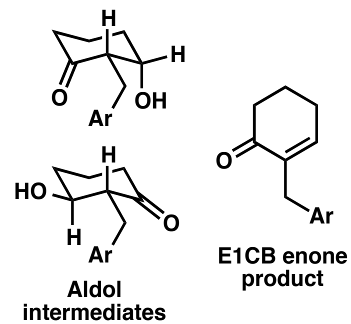
2. C–C and C–X bond forming reactions

2g) C–C Aldol reactions (*many with C. F. Barbas)

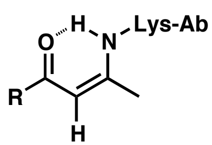
hapten | **substrate** | **products**

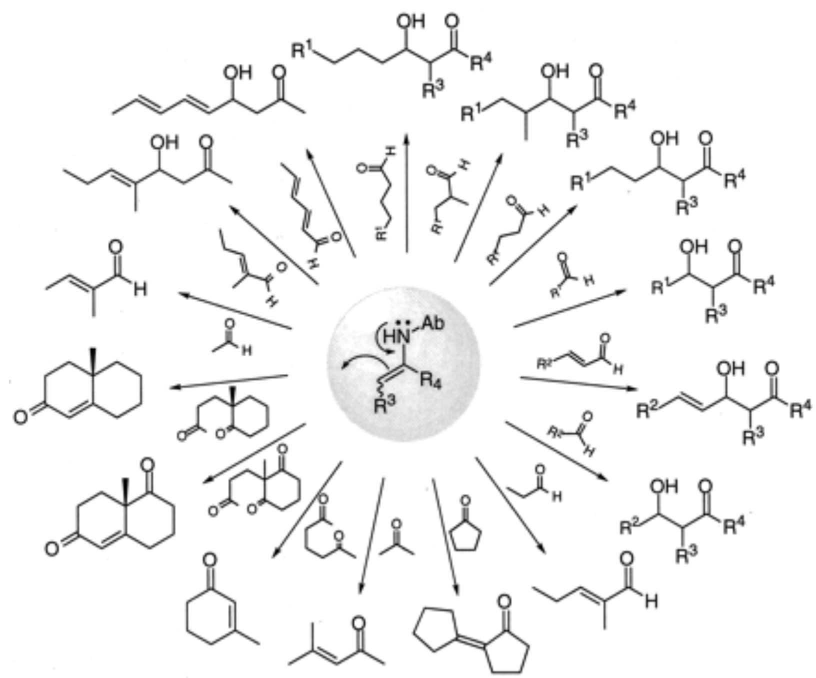






JACS **1995**,117, 9383





Science **1995**, 270, 1797
Science **1997**, 278, 2085
JACS **1998**, 120, 2768
PNAS **1998**, 95, 14603
TL **1999**, 40, 1437

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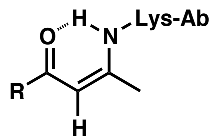
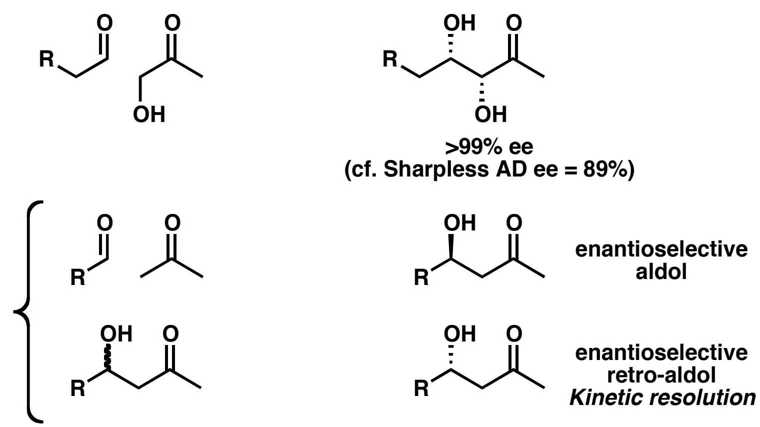
2. C–C and C–X bond forming reactions

2g) C–C Aldol reactions (*many with C. F. Barbas)

hapten **substrate** **products**

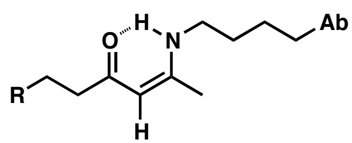


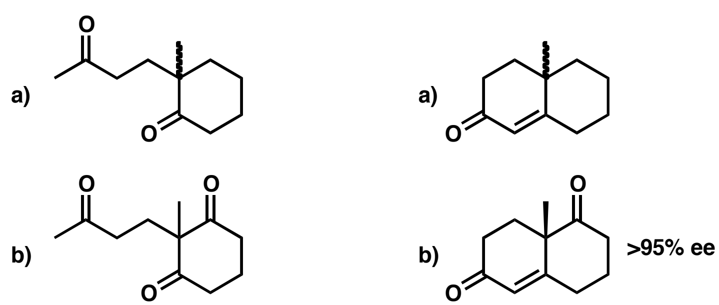


CEJ **1998**, 4, 881

ACIE **1998**, 37, 2481
ACIE **1999**, 38, 3738
JACS **1999**, 121, 7283
CEJ **2000**, 6, 2772
CEJ **2001**, 7, 1691
CBC **2001**, 2, 656

2h) C–C Robinson Annulation





JACS **1997**, 119, 8131
OL **1999**, 1, 598

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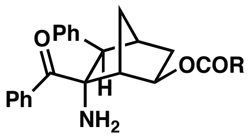
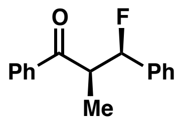
3. Other reactions

3a) Syn elimination

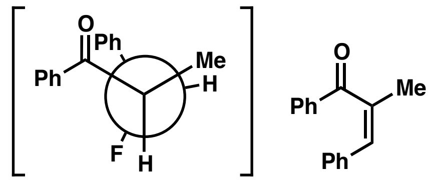
hapten



substrate



products



Eclipsed "disfavored" syn elimination

Z product (cis)

JACS **1994**, 116, 6013

3b) Oxidation of water

Antibodies, regardless of source or antigenic specificity, generate H_2O_2 from $^1O_2^*$ (singlet oxygen)

PNAS **2000**, 97, 10930

Science **2001**, 293, 1806