Masayuki Inoue and Total Synthesis of (+)-Ryanodine

Kuo Zhao

SED Group Meeting

3/15/16

Masayuki Inoue

- Born in February 14, 1971
- 1989-1993: The University of Tokyo, B.S, in Chemistry
- 1993-1998: The University of Tokyo, PhD. In Organic Chemistry, Advisor: Prof. Kazuo Tachibana
- Phd work: Synthetic studies toward Ciguatoxins
- 1998-2000: Sloan-Kettering Institute for Cancer Research, Postdoctoral fellow, Advisor: Samuel J. Danishefsky
- 2000-2004: Assistant professor, Associate professor (2004) at Tohoku University, work under Prof. Masahiro Hirama
- Achievement: total synthesis of cyclic polyether compounds Ciguatoxins produced by dinoflagellate
- 2007-present: Professor at the University of Tokyo

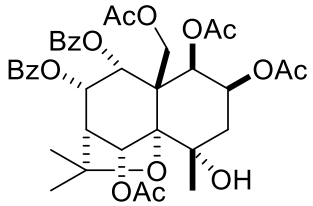
The man

Awards

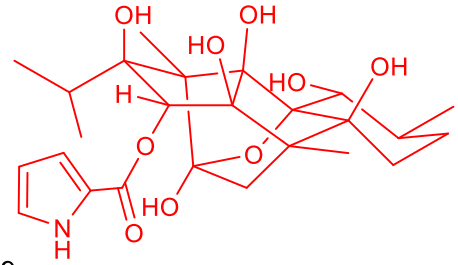
2001 Young Scientist's Research Award in Natural Product Chemistry
2001 Chugai Award in Synthetic Organic Chemistry, Japan
2004 First Merck-Banyu Lectureship Award
2004 The Chemical Society of Japan Award for Young Chemists
2005 Thieme Journal Award 2005
2007 Novartis Chemistry Lectureship 2008/2009
2008 Asian Core Program Lectureship Award 2009 (Singapore and Korea)
2009 Fifth JSPS PRIZE
2014 Mukaiyama Award Year 2014
2014 Fellow of the Royal Society of Chemistry

Inoue's Research Highlights
Total Synthesis

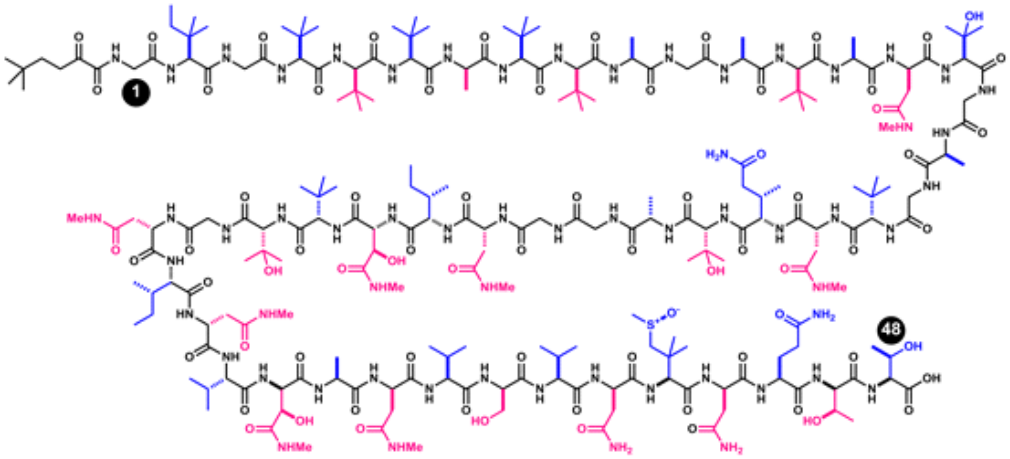
- Total synthesis of challenging terpenoids, polycyclic ether and polypeptide natural products.



(-)-hydroxyzinowol
J. Org. Chem., **2014**, *79*, 8835-8849

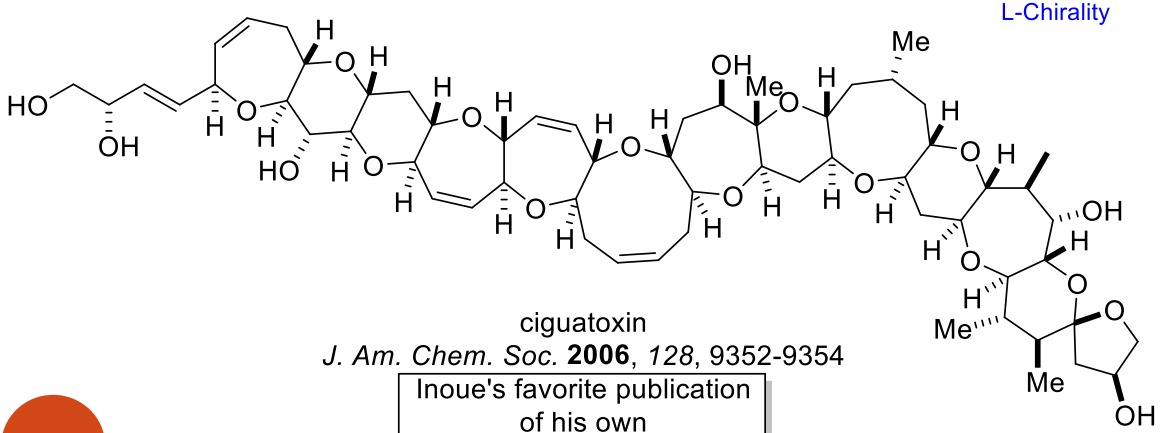


(+)-ryanodine
Chem. Eur. J. **2016**, *22*, 230-236
(asymetric total synthesis)



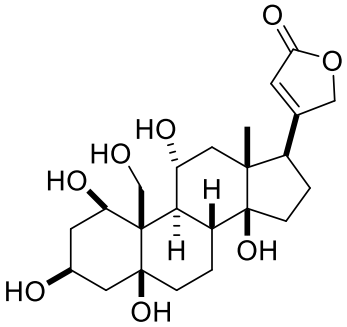
D-Chirality
L-Chirality

polytheonamide B
Nature Chem. **2010**, *2*, 280-285
48 amino acid units

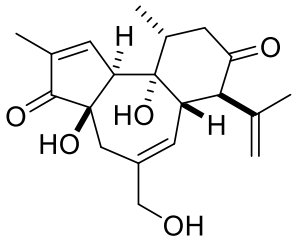


ciguatoxin
J. Am. Chem. Soc. **2006**, *128*, 9352-9354

Inoue's favorite publication of his own



ouabagenin
Chem. Sci., **2015**, *6*, 3383

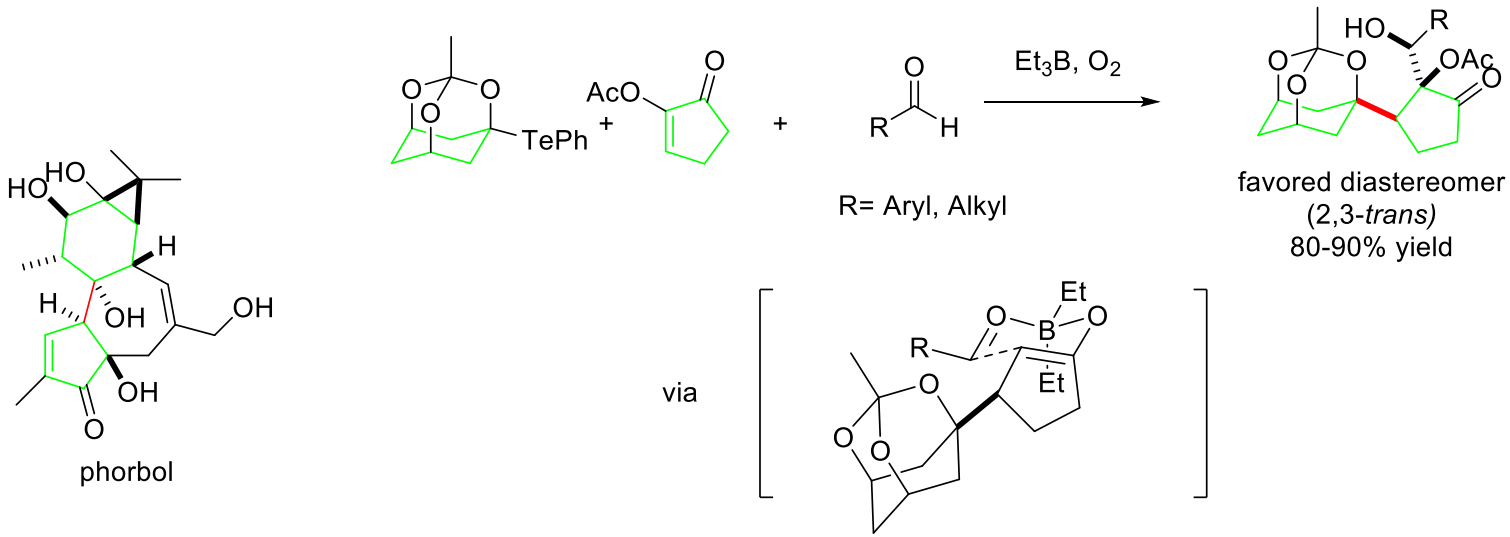


crotophorbolone
Angew. Chem. Int. Ed. **2015**, *54*, 14461-14457

Angew. Chem. Int. Ed. **2011**, 50, 9016

Inoue's Research Highlights Methodology design

1. Three-Component radical coupling;

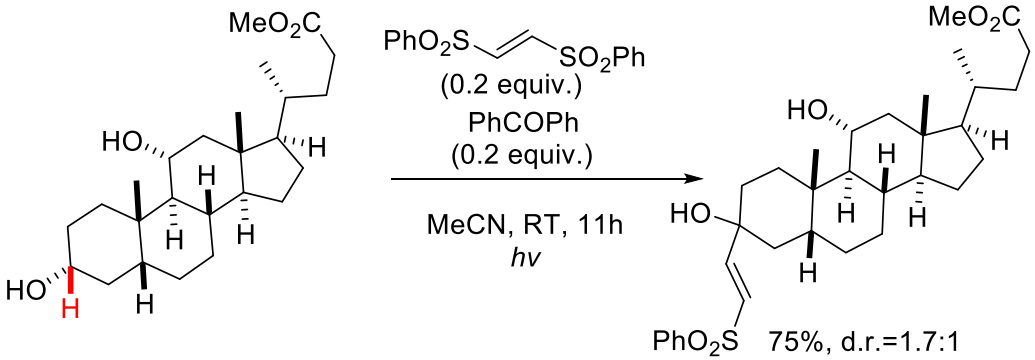


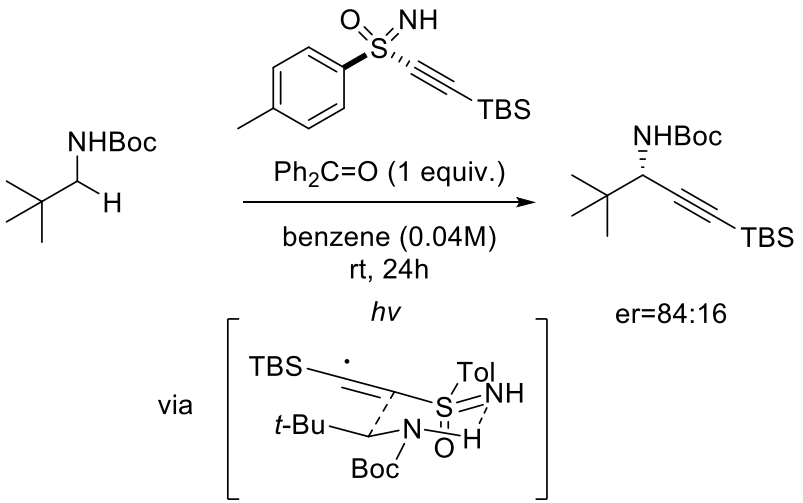
- Inoue group was able to prove that α-alkoxy bridgehead radical is robust and efficient in forming C-C bonds in hindered environment.
- Different combinations have also been developed by Inoue's group

Org. Lett., 2013, **15**, 5122-5125

Inoue's Research Highlights Methodology design

2. Radical/photochemical C-H activation;



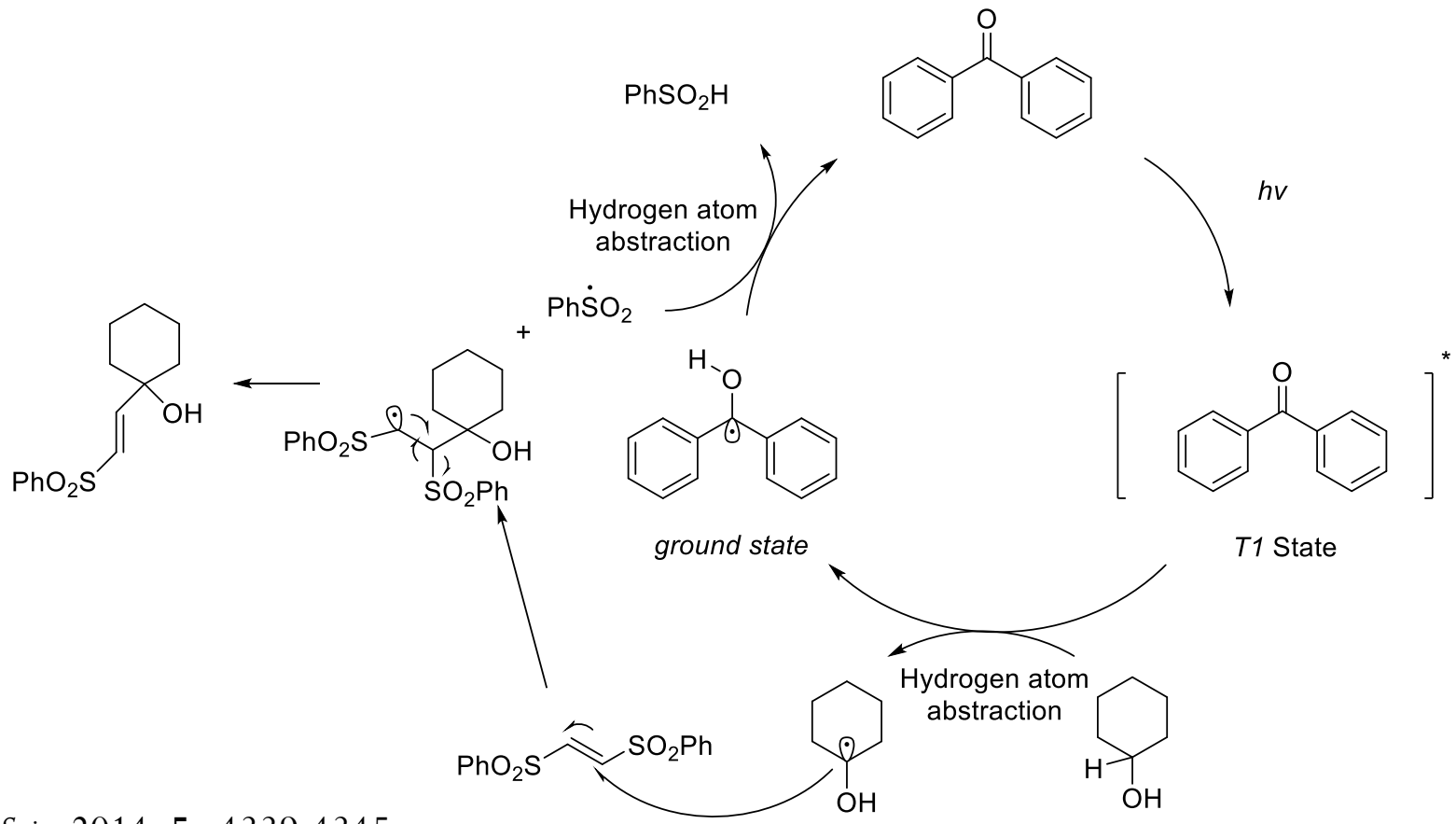


C(Sp3)-H Cynation, Alkynylation, 4-Pyridination, and Carbamoylation have been developed in similar manner.

Chem. Sci., **2014**, 5 , 4339-4345
Chem. Asian J. **2015**, 10, 120–123

Proposed Mechanism for Benzophenone Induced Photochemical C-H activation

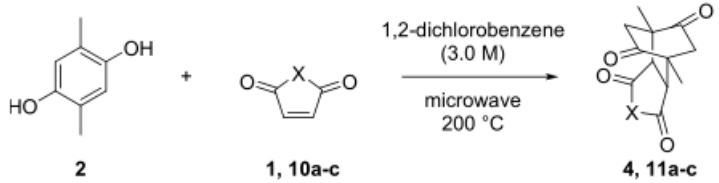
- Key idea: photoexcited carbonyl is a good reagent for hydrogen atom abstraction; one can view it as a *diradical* on carbon and oxygen.
- O-H high bond strength allows hydrogen atom abstraction to occur on oxygen
- C-H selectivity: least hindered and most nucleophilic C-H bond

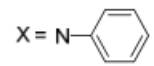
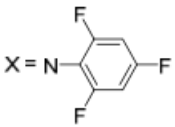
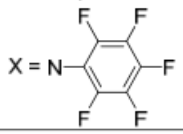


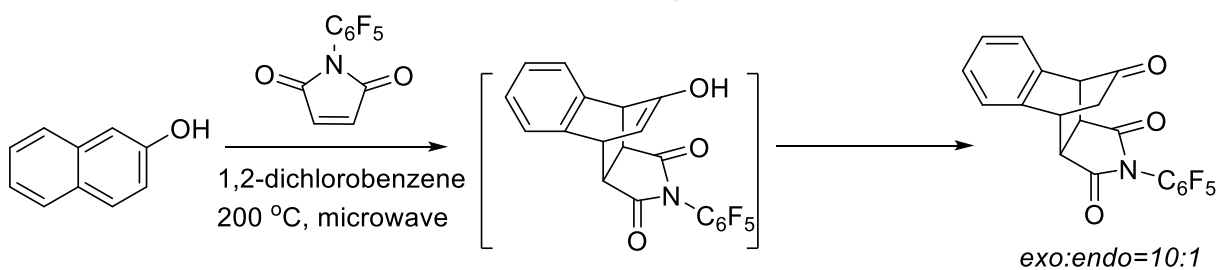
Chem. Sci., 2014, **5** , 4339-4345

Inoue's Research Highlights Methodology design

3. Dearomatizing Diels-Alder Reactions



entry	substituent	dienophile	product and yield
1	X = O	**1**	**4**: 16%
2	X = N– 	**10a**	**11a**: 23%
3	X = N– 	**10b**	**11b**: 37%
4	X = N– 	**10c**	**11c**: 40%



exo:endo=10:1
70%

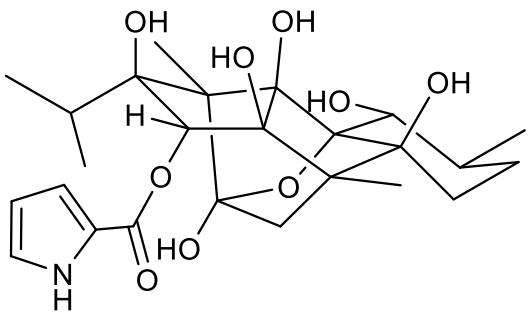
- The electron withdrawing nature of the dienophiles increase the yield
- *exo* 4+2 adduct is favored because of steric effect.
- Design for synthesis of ryanodol&ryanodine's core.

Heterocycles, **2015**, *90*, 659-672

(+)-Ryanodine: A Brief Summary

- One of ryanoids-natural products found from the genus *Ryania*. This particular one was separated from *Ryania speciosa Vahl* 's stem and root material in 1948. Its structure was proposed *almost* correctly in 1967 and was finally determined in 1968 by XRD.

Ryania Speciosa Vahl
(By A. Hernandez)



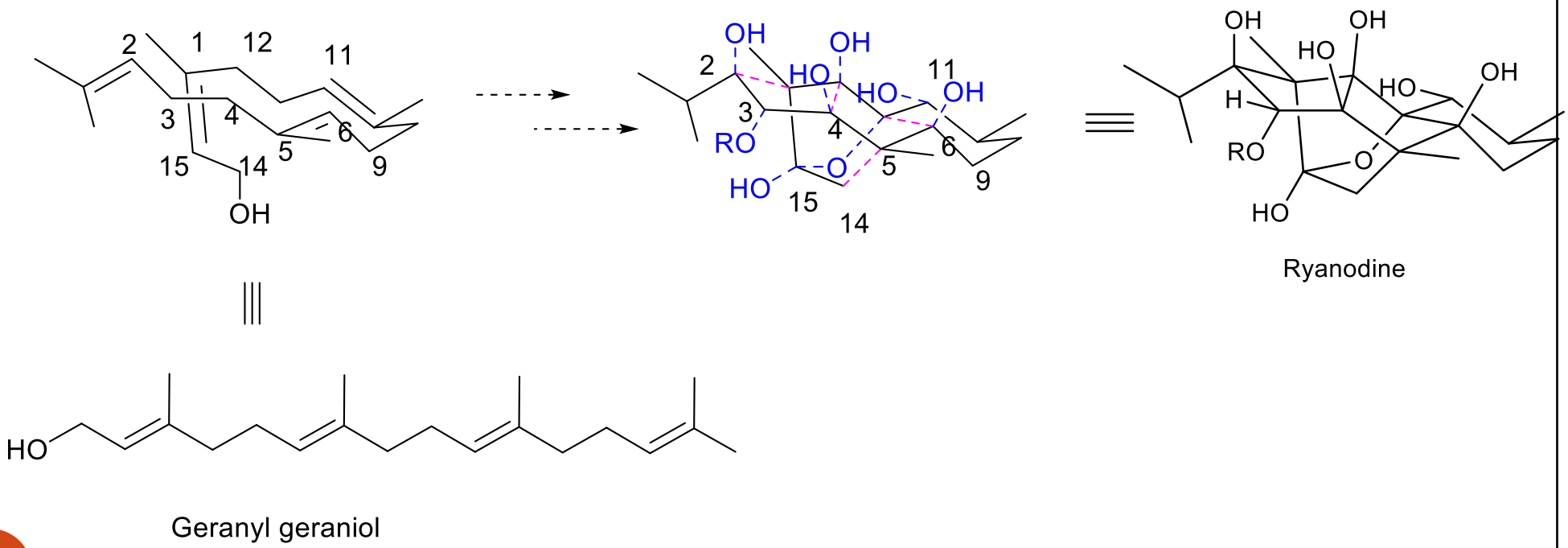
(+)-ryanodine

- Ryanodine is found to bind strongly with *Ryanodine Receptors* (RyRs) in open form and causes RyRs to close (RyRs control the release of Ca2+ to muscle/neuron cells) , which could lead to paralysis at micromolar concentration. Ryanodine is a strong toxic and insecticide for this reason(work for both mammals and insects) .

Tetrahedron Lett. **1967**, *8*, 221
J. Am. Chem. Soc., **1948**, *70*, 3086–3088
Can. J. Chem. **1968**, *46*, 795

Proposed Biosynthsis

- No studies on the biosynthesis of ryanodine have been reported until today.
- The ryanoid core contains 20 carbon atoms, which can be classified as highly oxygenated polycyclic diterpenoids.
- Diterpenoid Geranyl geraniol is proposed to be the reasonable start point of this core construction, but there is no evidence.



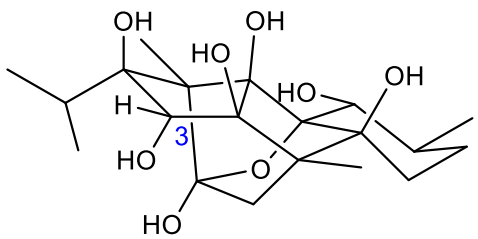
Graeber, J. K. *Yale University Graduate Thesis* **2003**

Challenges in Synthesis of Ryanodine

•Ryanodine contains a complex, cage shaped pentacylic core with 11 contiguous stereocenters and many sensitive FGs (5 hydroxyls, 1 hemiketal)

•Ryanodol, the precursor of Ryanodine, has been successfully synthesized since 1979 (although to date only two groups have achieved its total synthesis) However, it took another ***36*** years for the esterification to be accomplished!

•3-*epi*-ryanodol is what has been recently discovered to be the *natural ryanodol* by Inoue's group!

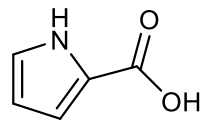


(+)-ryanodol (synthetic)

Can. J. Chem. **1979**, *57*, 3348
(By Pierre Deslongchamps et al.)

J. Am. Chem. Soc., **2014**, *136*, 5916-5919
(By M. Inoue et al.)

Chem. Eur. J. **2016**, *22*, 230-236
(By M.Inoue et al.)

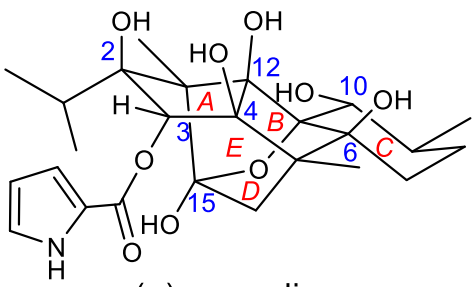


C3 esterification

Selectivity?
How?

KOH, EtOH

by K. Wiesner
Can. J. Chem. **1951**, *29*, 905



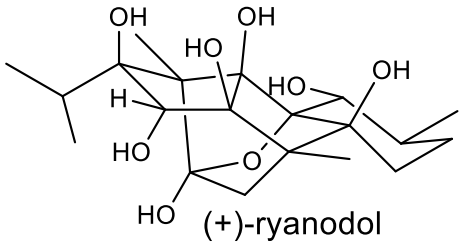
(+)-ryanodine

Chem. Eur. J. **2016**, *22*, 230-236
(By M.Inoue et al.
first successful total synthesis of ryanodine ever)

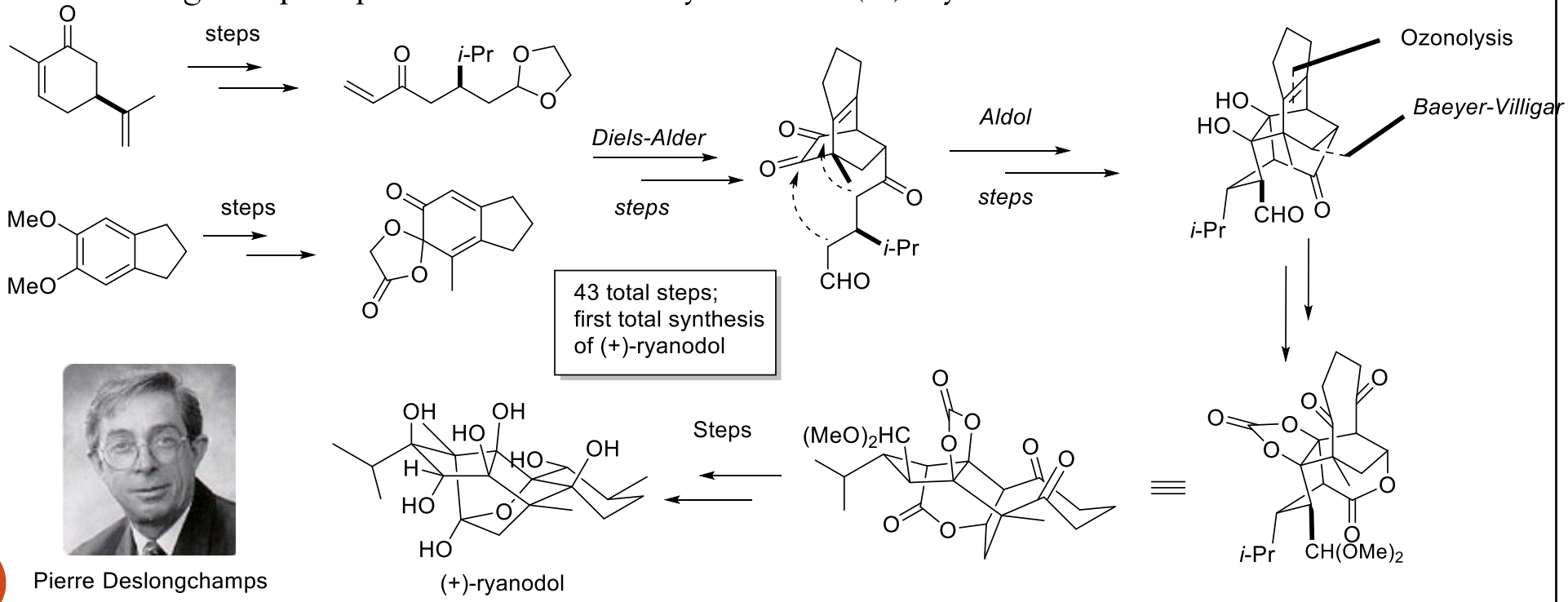
Angew. Chem. **2016**, *128*, 2539–2543

Previous Synthetic Effort toward Ryanodol and Ryanodine

- Ryanodol is the key precursor of Ryanodine. It shares all structural features with Ryanodine except for the α-pyrrole ester.



- P. Deslongchamps reported the first total synthesis of (+)-Ryanodol in 1979.

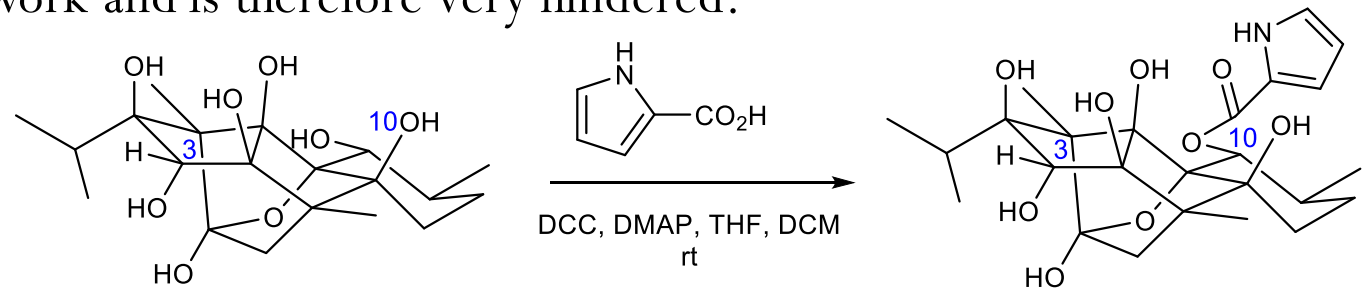


Pierre Deslongchamps

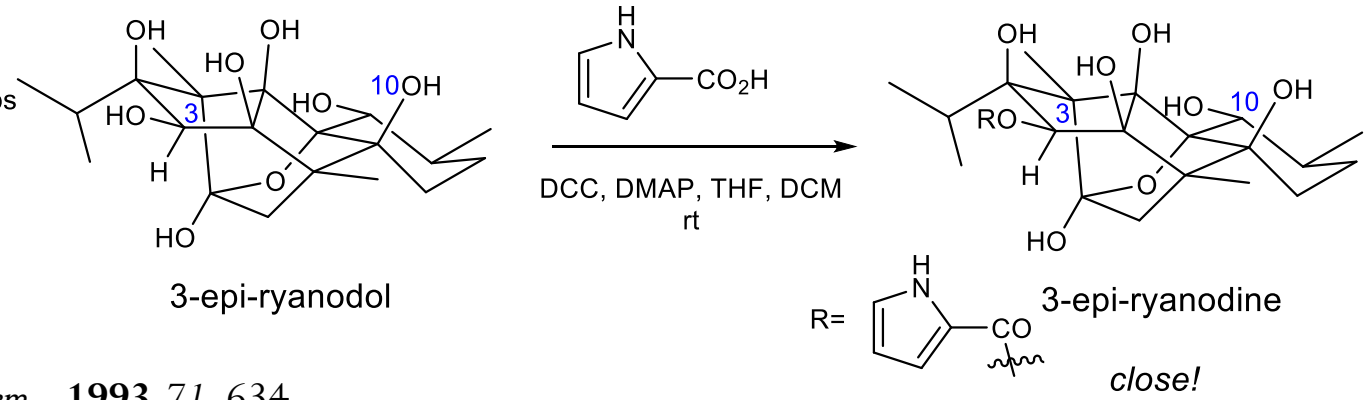
Can. J. Chem. **1979**, *57*, 3348

Previous Synthetic Effort toward Ryanodol and Ryanodine

- Attempt toward the synthesis of ryanodine by Deslongchamps was fruitless
- Innate regioselectivity of esterification site is C10 not C3 on ryanodol!
- 3-epi-ryanodol gives C3 ester. Ryanodol's C3 alcohol is inside of concave carbon framework and is therefore very hindered.



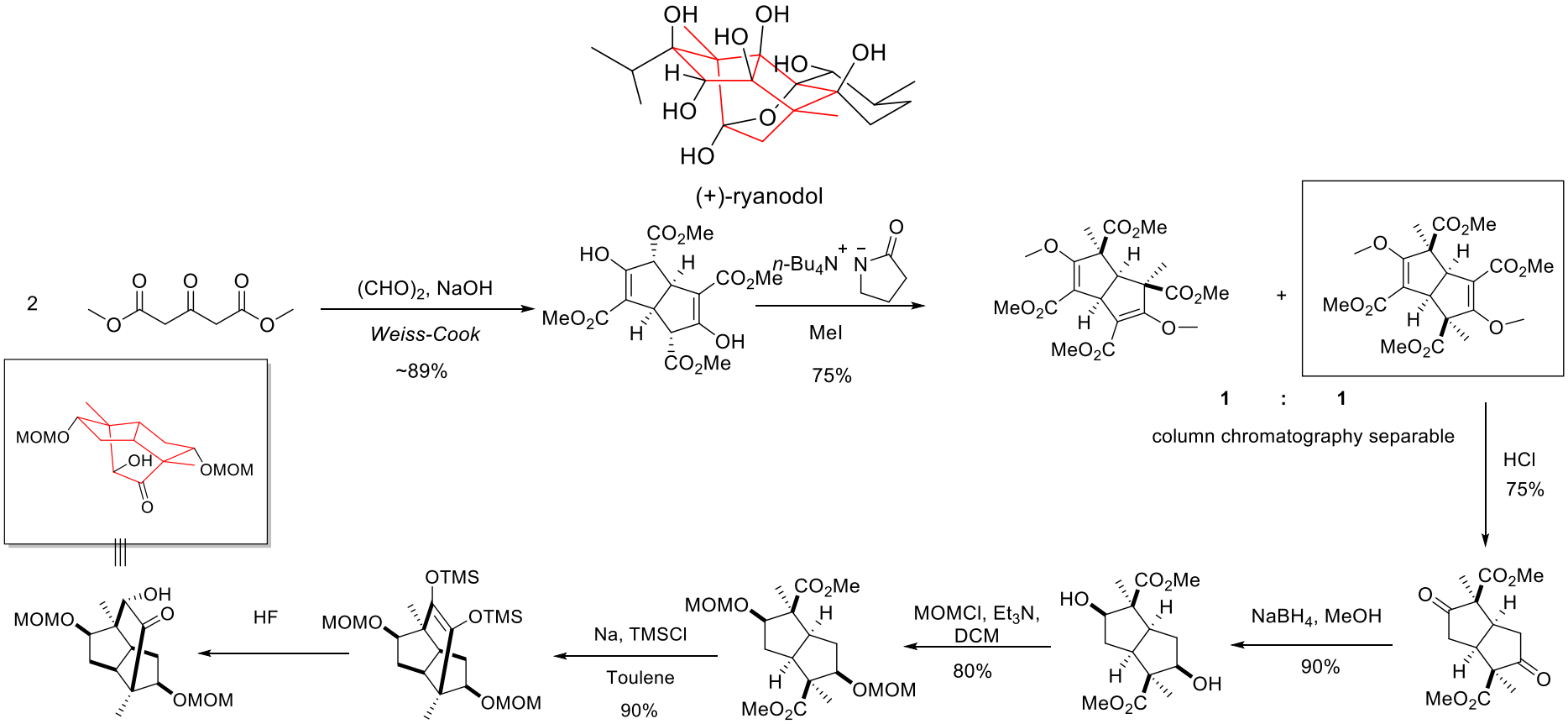
Pierre Deslongchamps



Can. J. Chem., **1993**, *71*, 634

Previous Synthetic Effort toward Ryanodol and Ryanodine

- Scott Sieburth's group attempted another synthesis in 1987 and was able to generate a clever route toward ryanodol's C2 symmetric core.

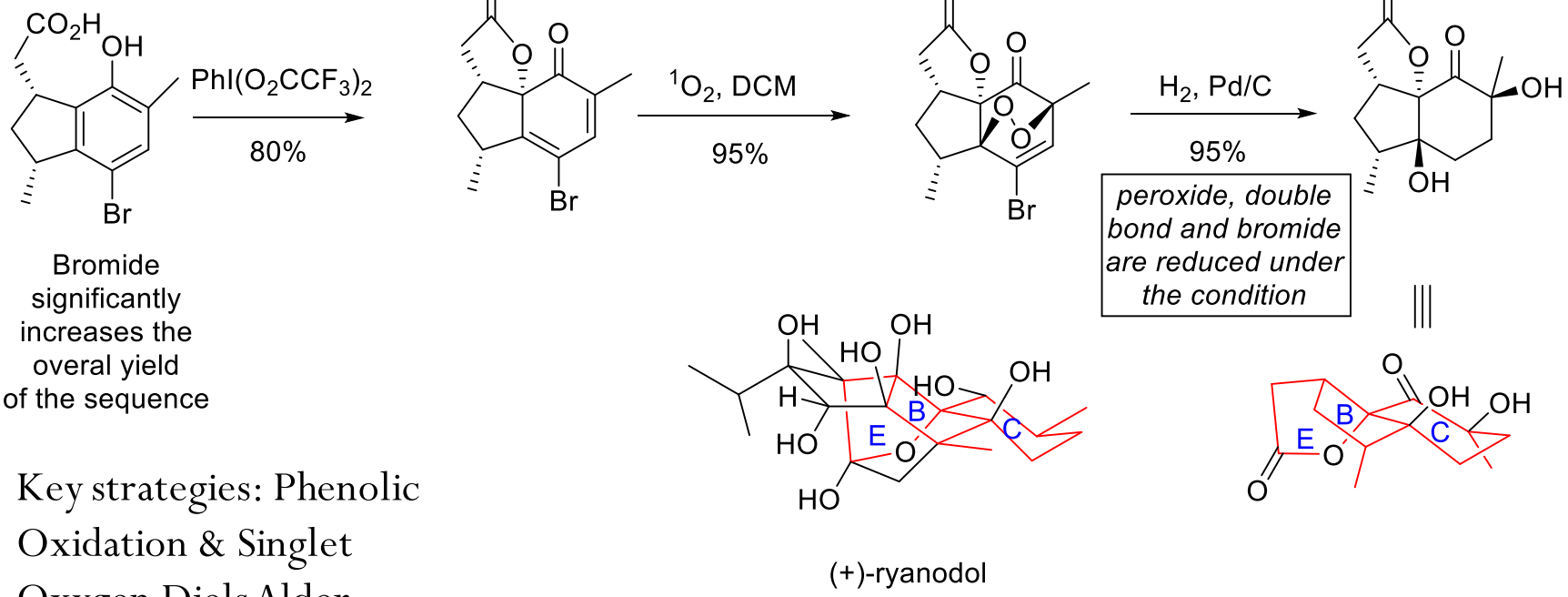


Tetrahedron Lett. **1994**, *35*, 8127
Org. Synth. **1986**, *64*, 27

Previous Synthetic Effort toward Ryanodol and Ryanodine

- John Wood's group reported a synthetic study of Ryanodol's BCE ring in 2003. However, due to lack of necessary functionalities, this route could not lead to the target molecule

John Wood



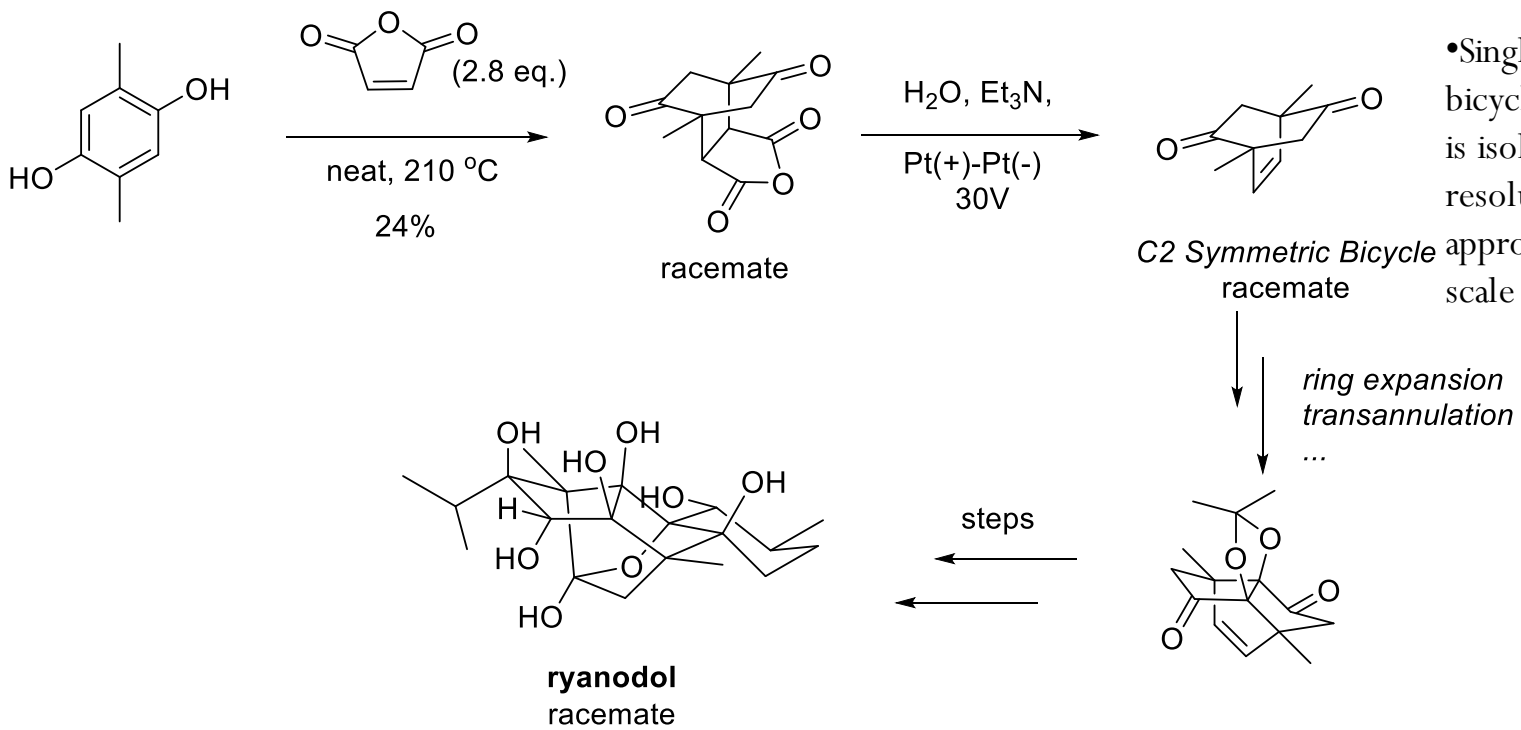
Bromide significantly increases the overal yield of the sequence

Key strategies: Phenolic Oxidation & Singlet Oxygen Diels Alder

Tetrahedron., **2003**, *59*, 8855

Previous Synthetic Efford toward Ryanodol and Ryanodine

- In 2014, M.Inoue's group published a racemic total synthesis of ryanodol.

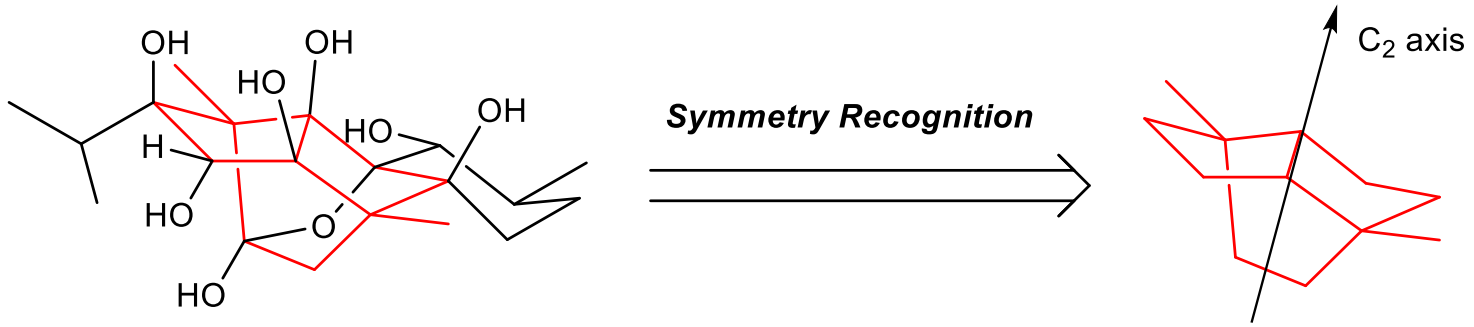


•Single enantiomer of this bicycle intermediate's derivative is isolatable via enzyme's kinetic resolution from a different approach, but it is too hard to scale up.

J.Am. Chem. Soc. **2014**, *136*, 5916−5919
Heterocycles **2010**, *82*, 491

Inoue's Symmetry-Driven Strategies Toward Synthesis of (+)-Ryanodol and (+)-Ryanodine

- Inoue noticed that the core of ryanodol has a C2 symmetric tricyclic component.



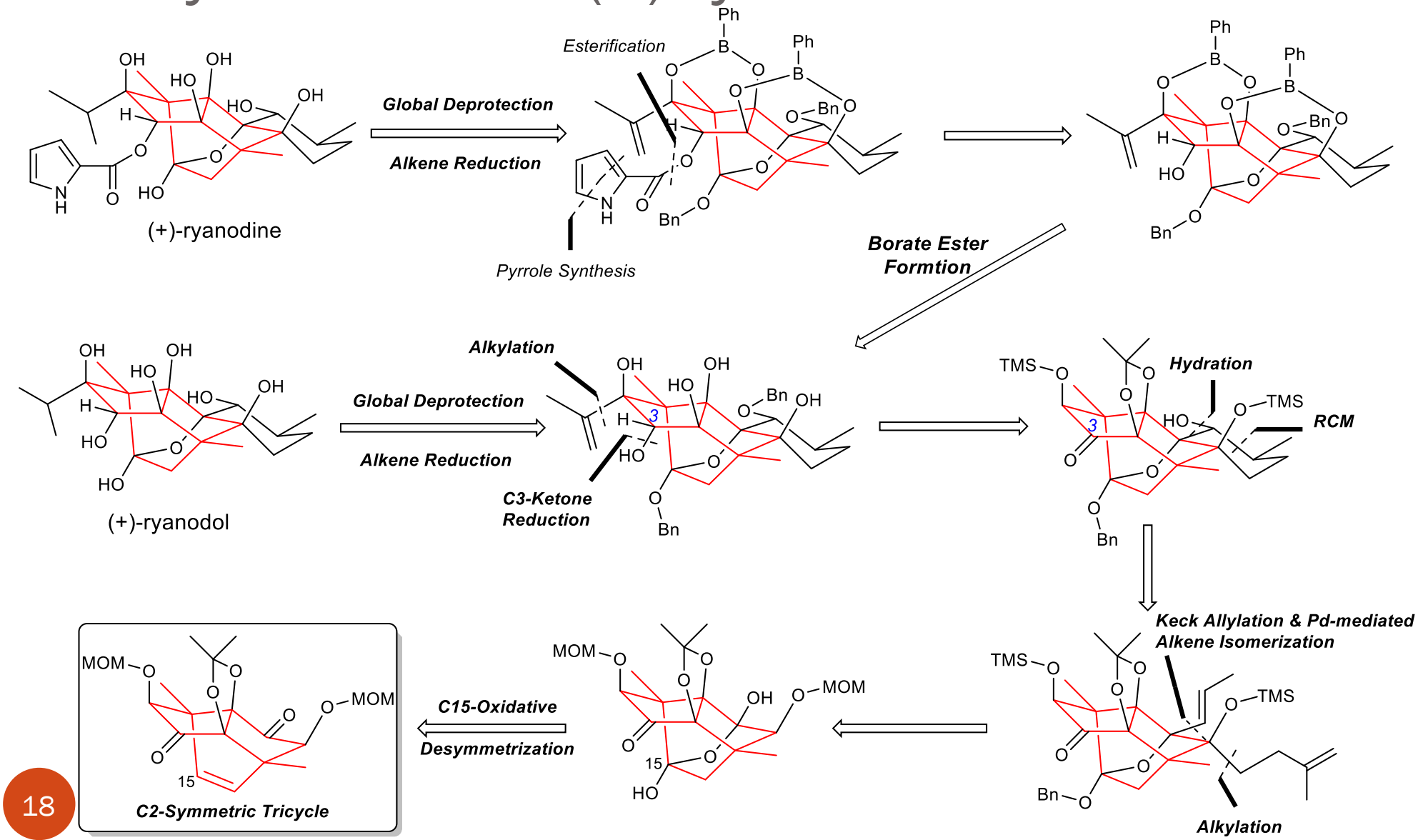
(+)-ryanodol

C_2 Symmetric Tricycle

- Advantages of *C2* symmetry core strategy: allow *two* reaction to run on *two* functionalities in ***one*** synthetic step—reduce step number!

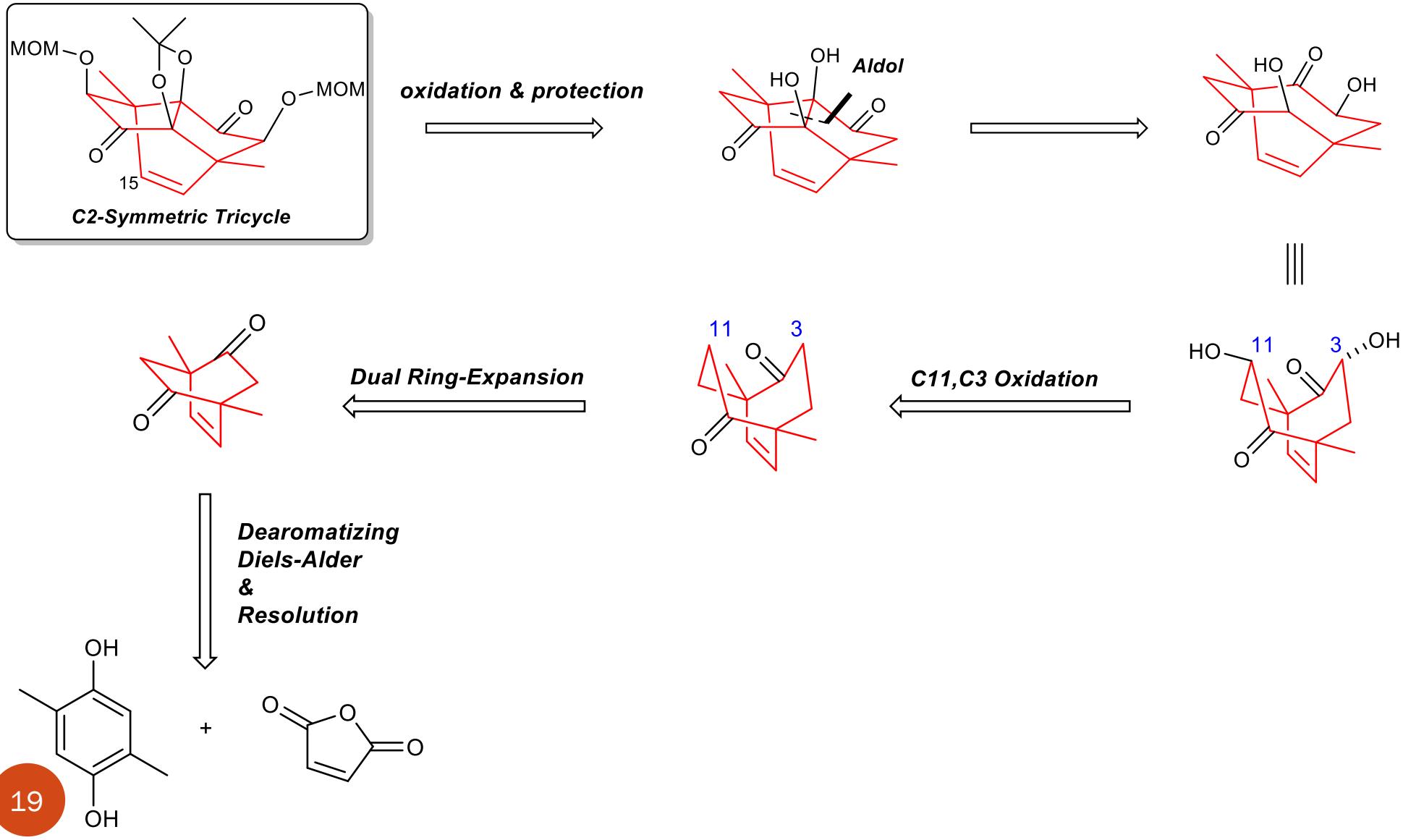
J. Am. Chem. Soc. **2014**, *136*, 5916−5919

Inoue's Retrosynthetic Analysis of (+)-Ryanodol and (+)Ryanodine

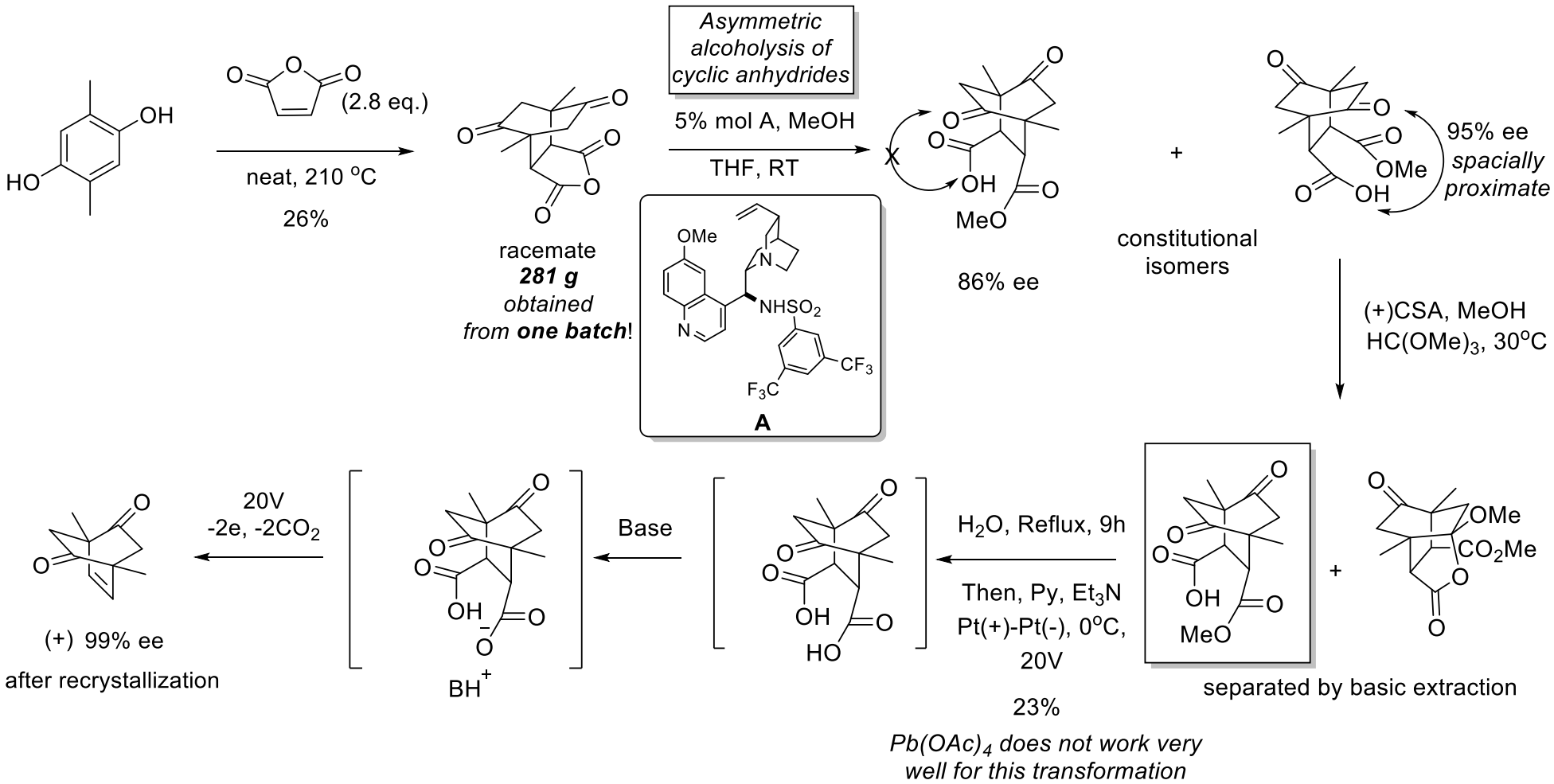


Chem. Eur. J. **2016**, *22*, 222 – 229
Chem. Eur. J. **2016**, *22*, 230 – 236

Inoue's Retrosynthetic Analysis of (+)-Ryanodol and (+)Ryanodine

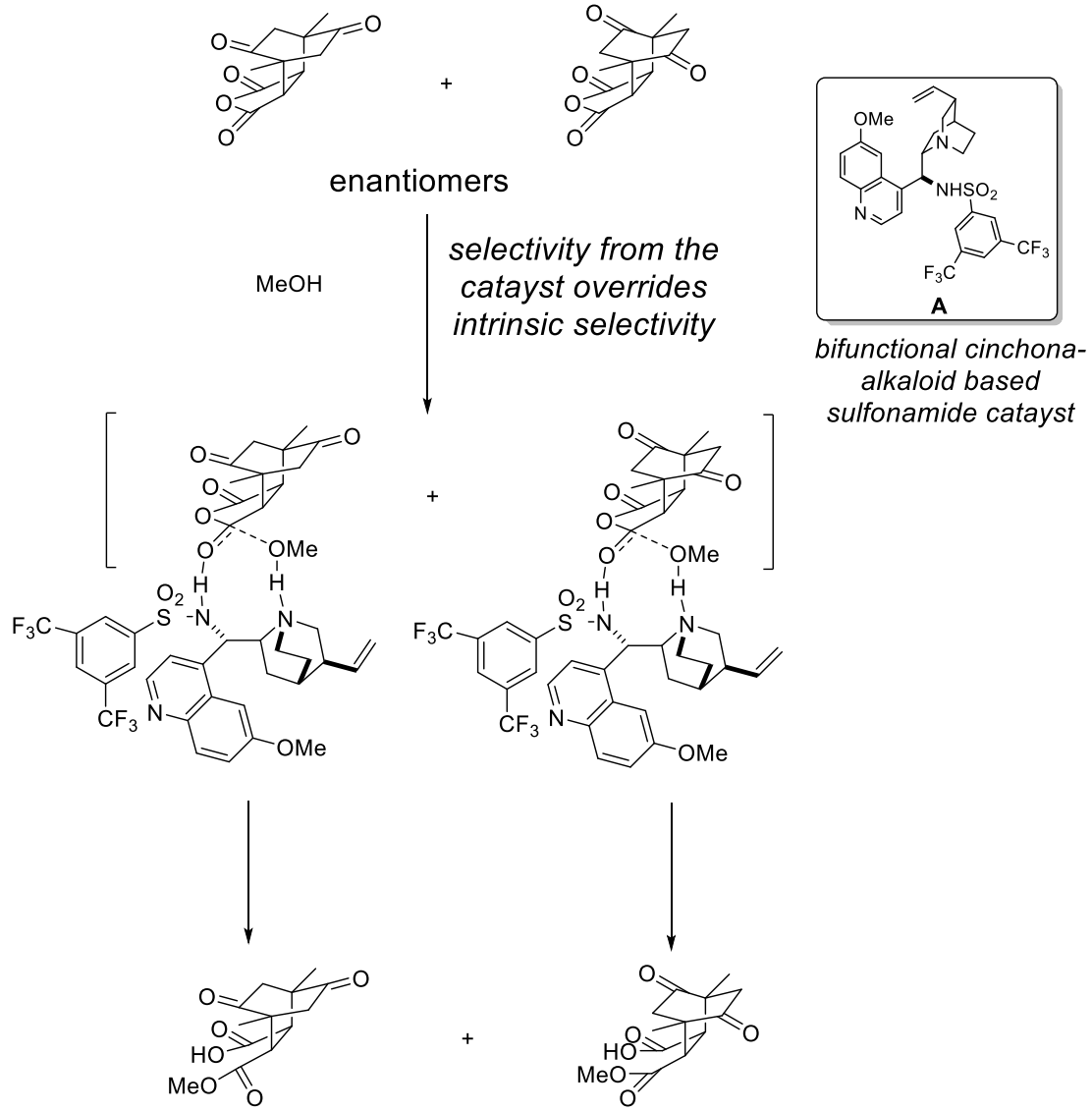


Synthesis of Enantiopure C2-Symmetry Bicycle Precursor

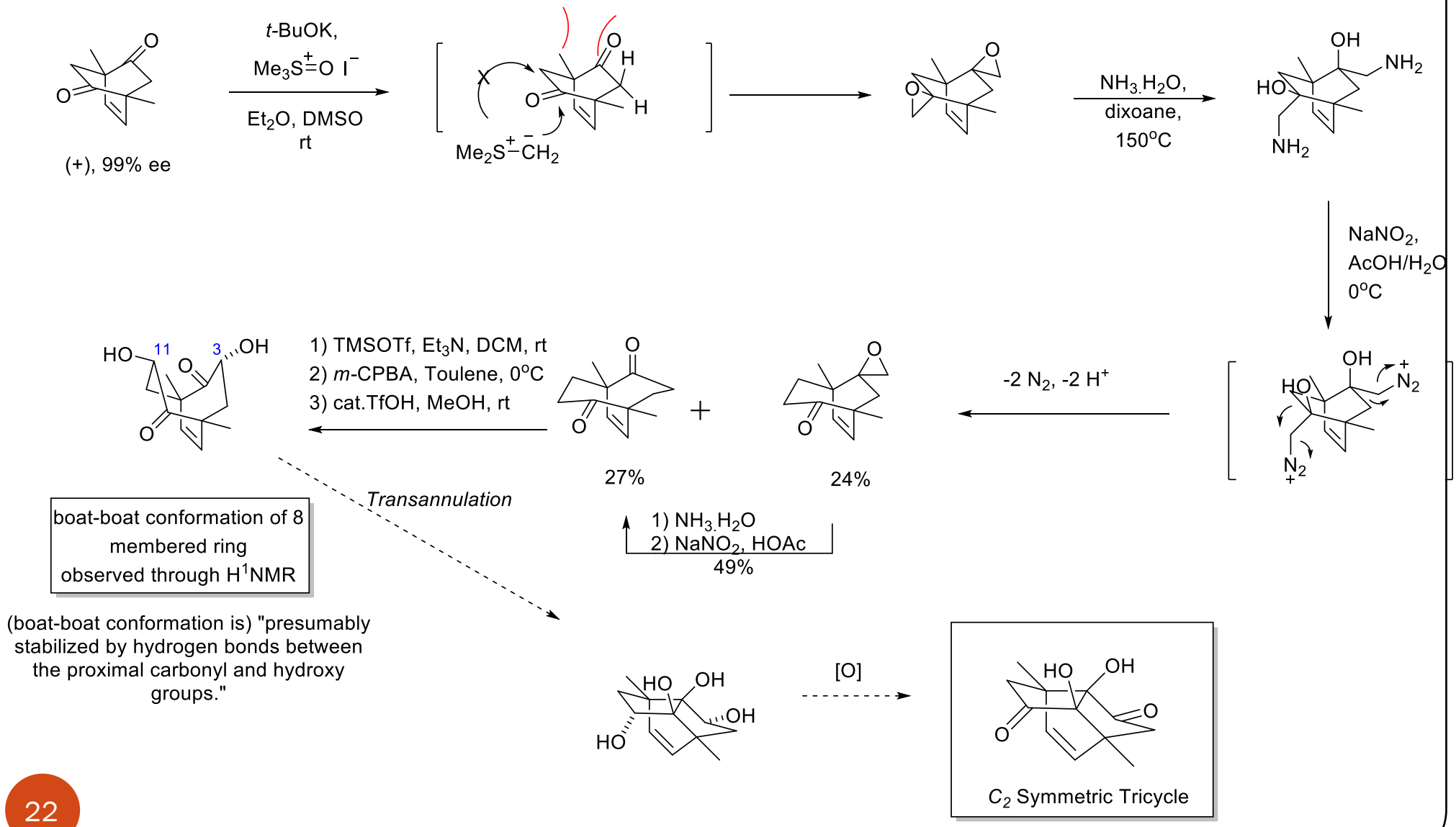


- Four-step sequence. Low yield is compensated by easy separation&purifcation and very large scale for each step (ten to hundred-gram scale).

Mechanism of Asymmetric Methanolysis of Cyclic Anhydride (Resolution)



Synthesis of *C2 Symmetric* Tricyclic Intermediate

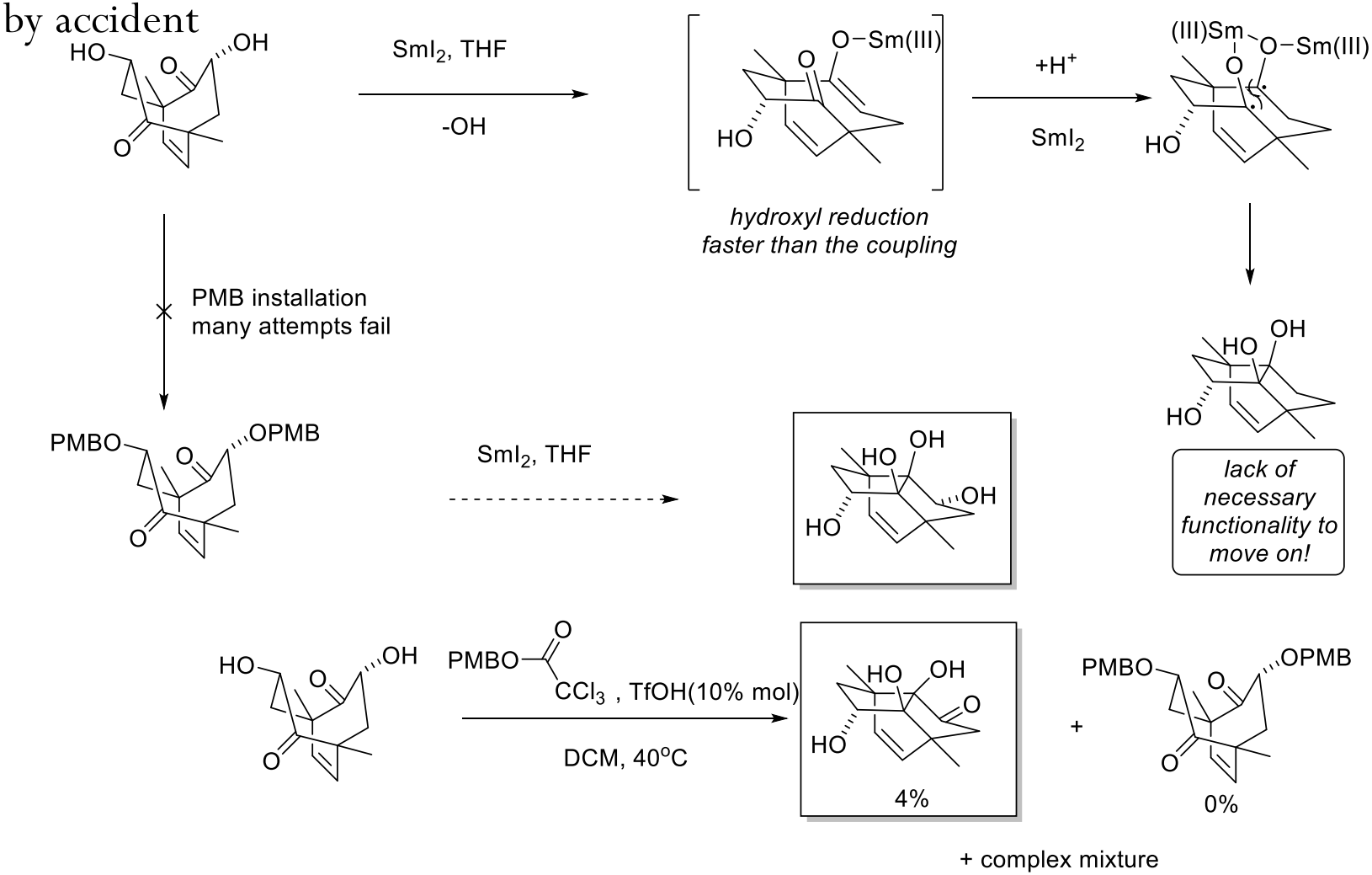


boat-boat conformation of 8 membered ring observed through H^1NMR

(boat-boat conformation is) "presumably stabilized by hydrogen bonds between the proximal carbonyl and hydroxy groups."

Study on Transannulation

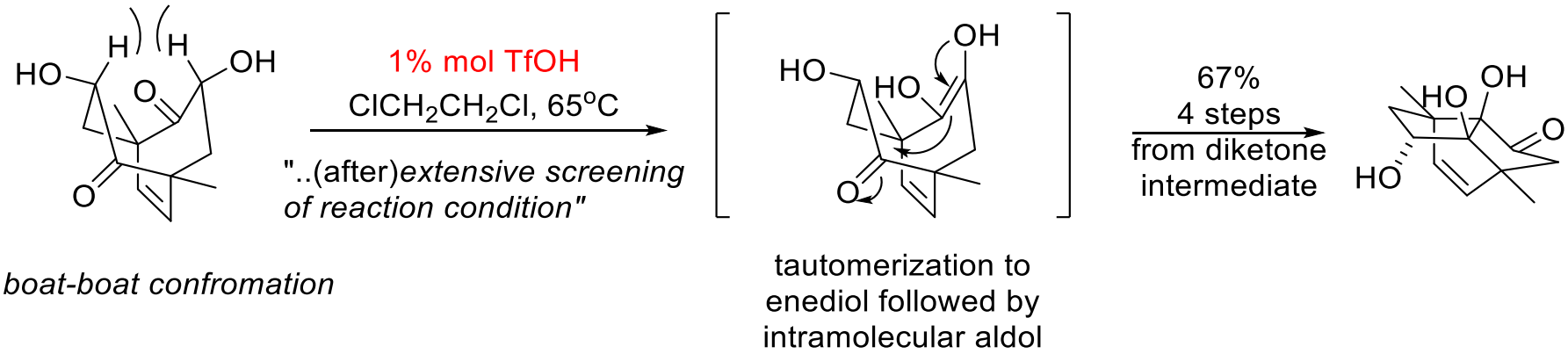
- Intramolecualr reductive coupling of the diketone via SmI2 indeed occured but over reduction happened.
- In an attempt to install PMB on hydroxyls, they discovered the target transannulation by accident



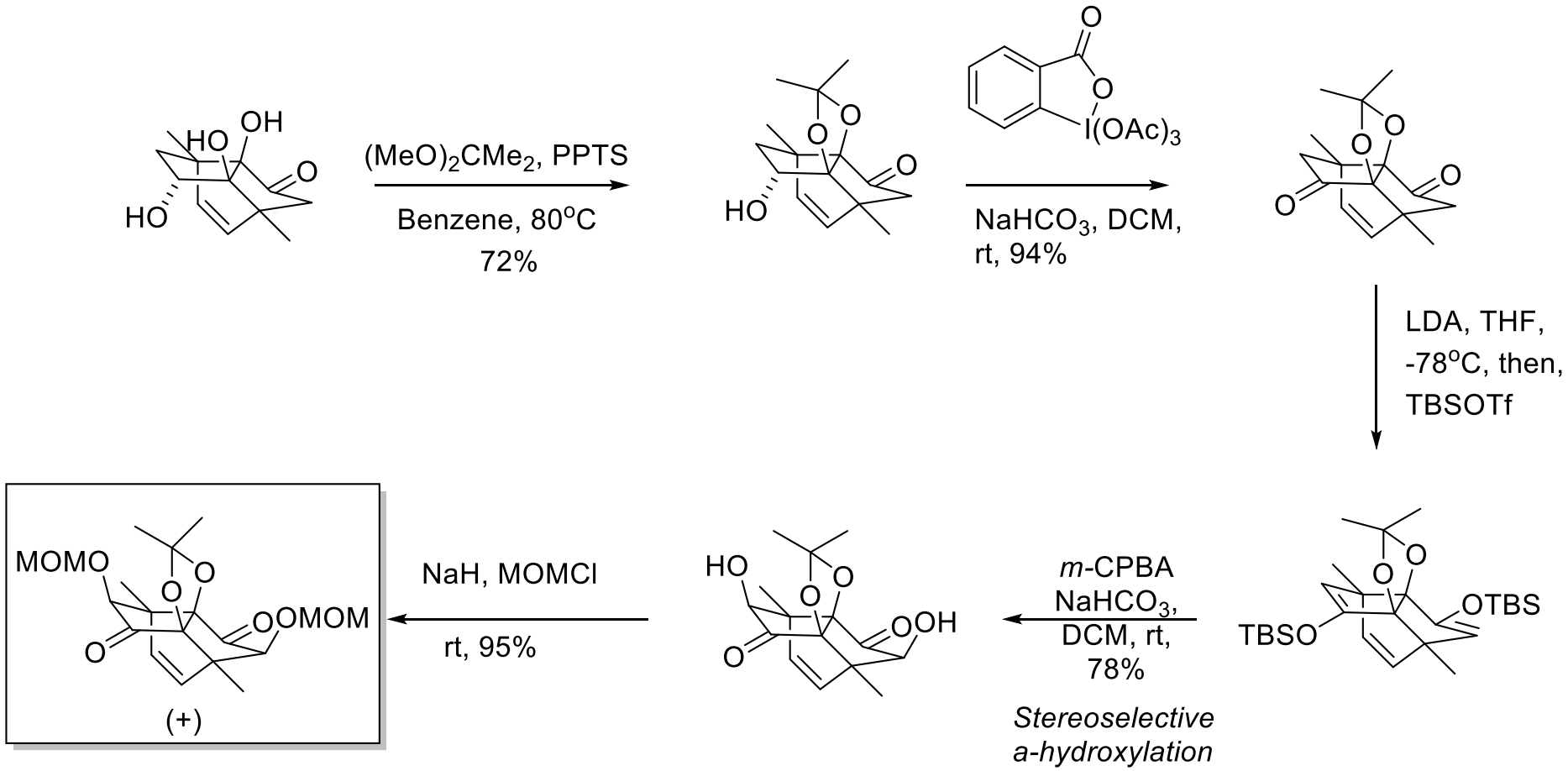
Study on Transannulation

- TfOH is discovered to be an effective catalyst for this transannulation.
- Enolization is considered to easily happen due to releasing transannular strain created by boat-boat conformation.

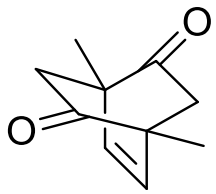
transannular strain leads to easy enolization



Synthesis of *C2 Symmetric* Tricyclic Intermediate

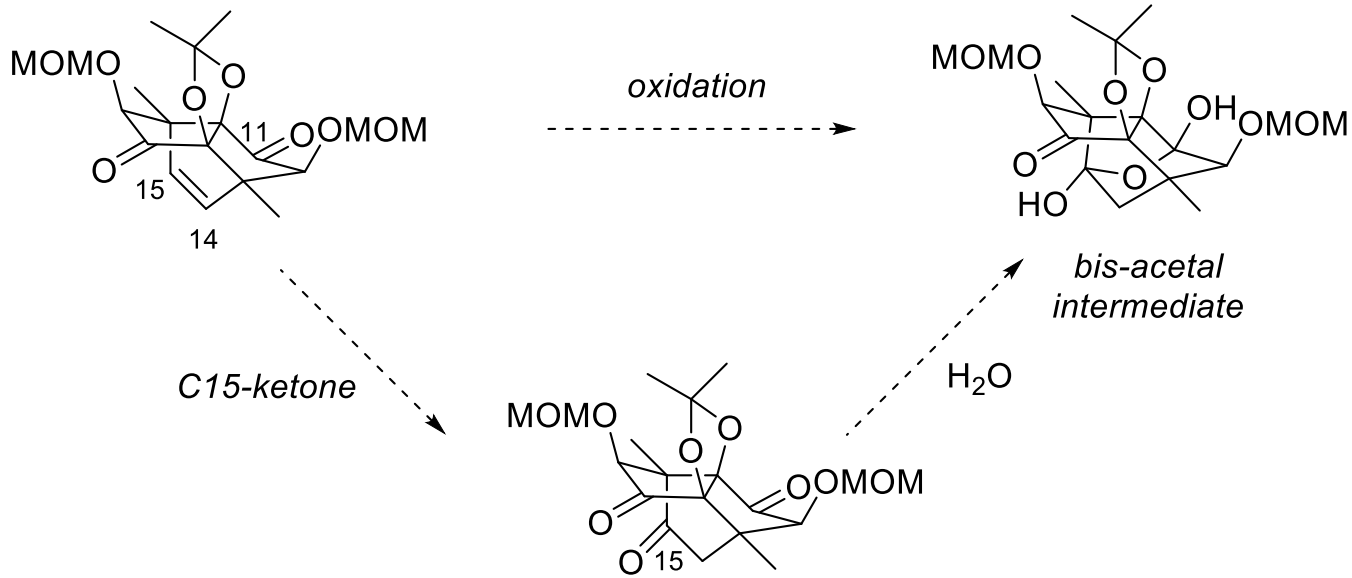


Summery: double tandem functionalizations on *C2* symmetric deketone significantly increased the efficiency of synthesis!



Construction of ABDE Rings of Ryanodine: Initial Attempt

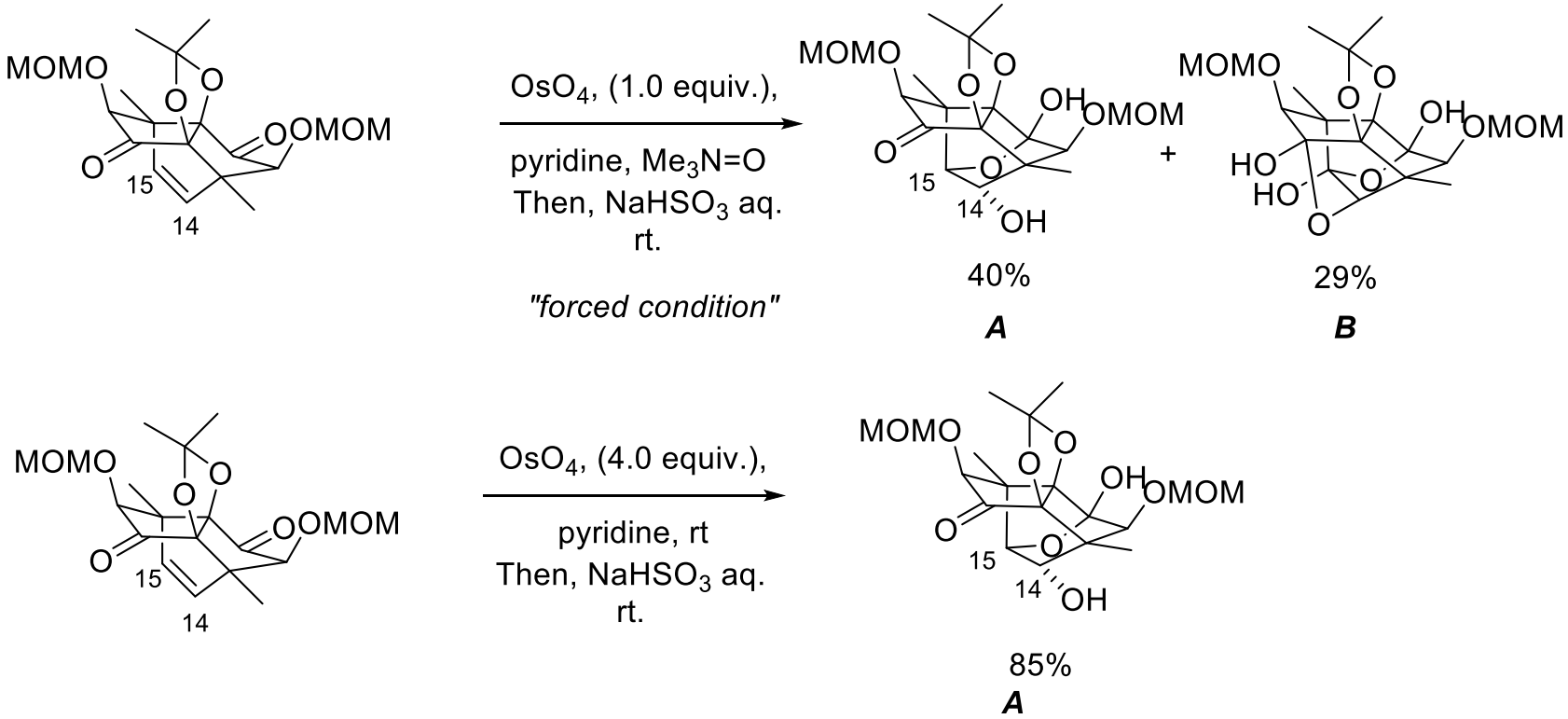
- First thought: C15-olefin converted into C15-ketone is easy because the corresponding bis-hemiacetal(C11,C15) is very stable.
- Reality: highly challenging due to hindered environment. Hydroboration–oxidation, oxymercuration–demercuration and iodohydrin formation all failed to introduce C15-ketone or bis-hemiacetal.



a note on symmetry: C14 is equal to C15 here due to intrinsic C_2 symmetry

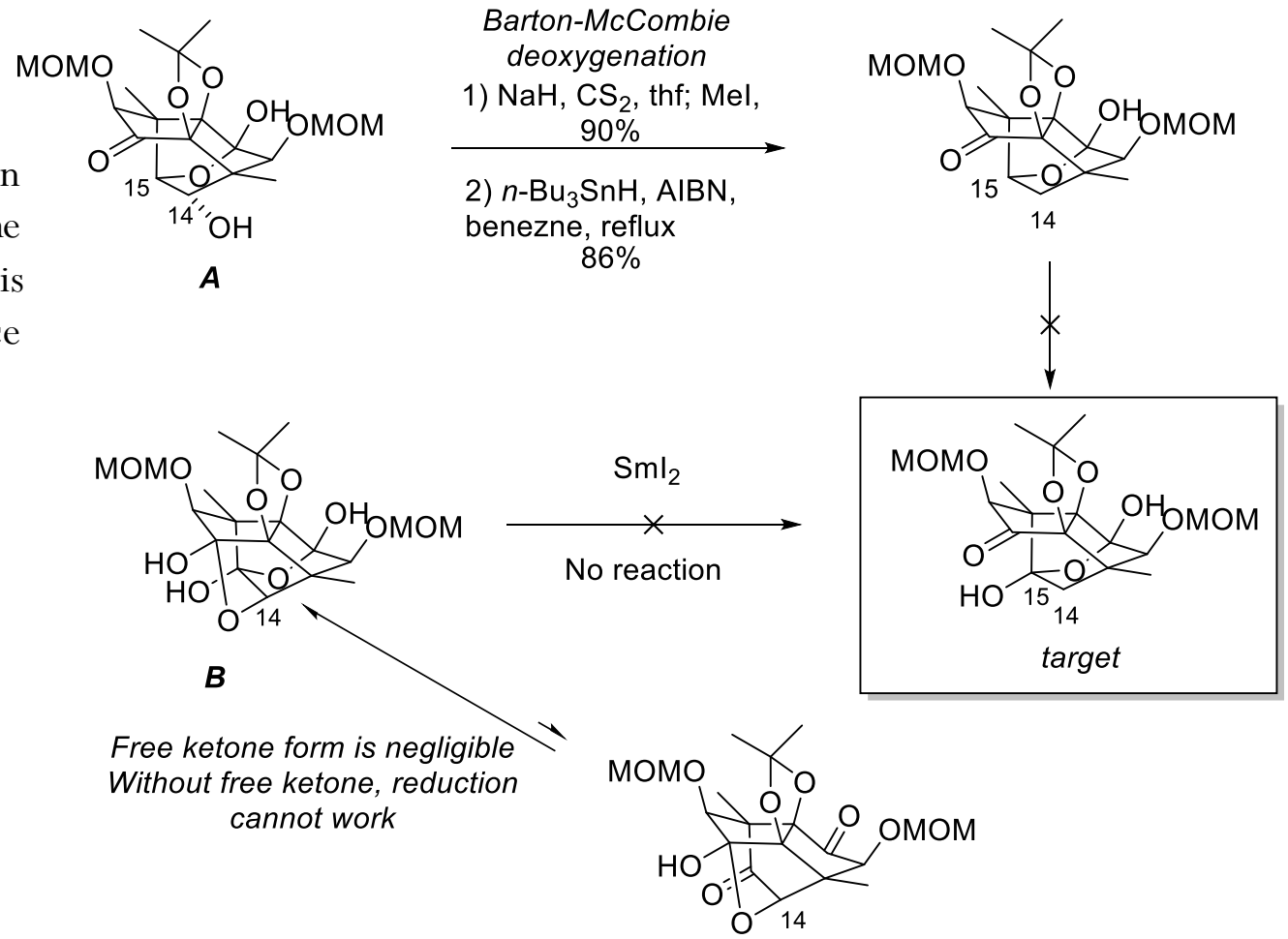
Construction of ABDE Rings of (+)-Ryanodine: Initial Attempt

- Osmylation under forced condition lead to oxidation of C15 and C14 (product A) and an over-oxidized byproduct B.
- When milder condition was used, A is the only product.



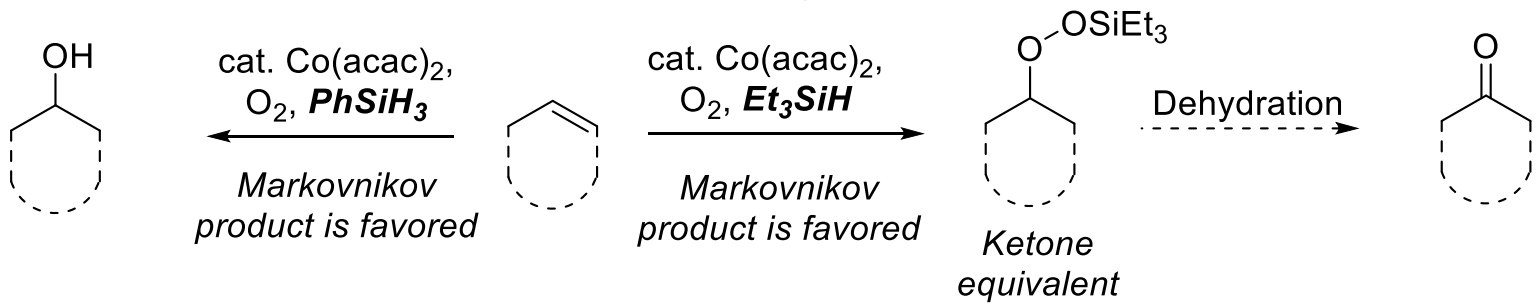
Another Deadend...

- For intermediate A, C14 reduction and C15 oxidation would lead to the target. However, C15 oxidation is not accessible due to "the presence of more reactive C-H bonds."
- For intermediate B, C14 hydroxyl reduction would lead to the target. It did not work either.



Breakthrough: Development of New Methodology

- The only way to solve this problem is to directly introduce a carbonyl on C15, not an C15 hydroxyl; C14 oxidation should also be avoided.
- Mukaiyama-Isayama Hydroperoxysilylation of alkene was noticed by Inoue's group. The resulting peroxysilane is a ketone surrogate. Elimination could lead to ketone.



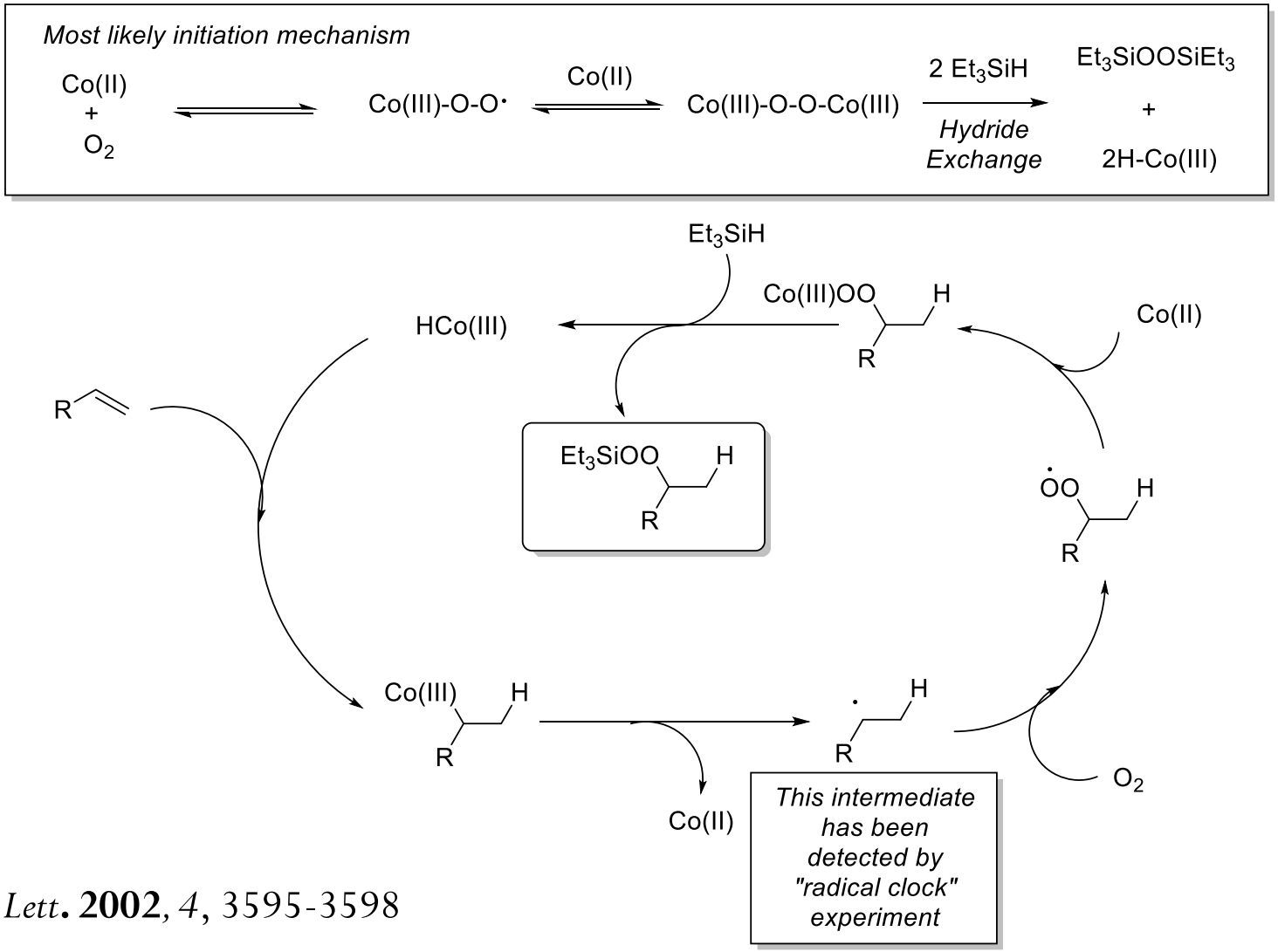
Teruaki Mukaiyama

Shigeru Isayama

Chem. Lett. **1989**, 1071

Mechanism of Cobalt Catalyzed Hydroperoxysilylation on Olefin

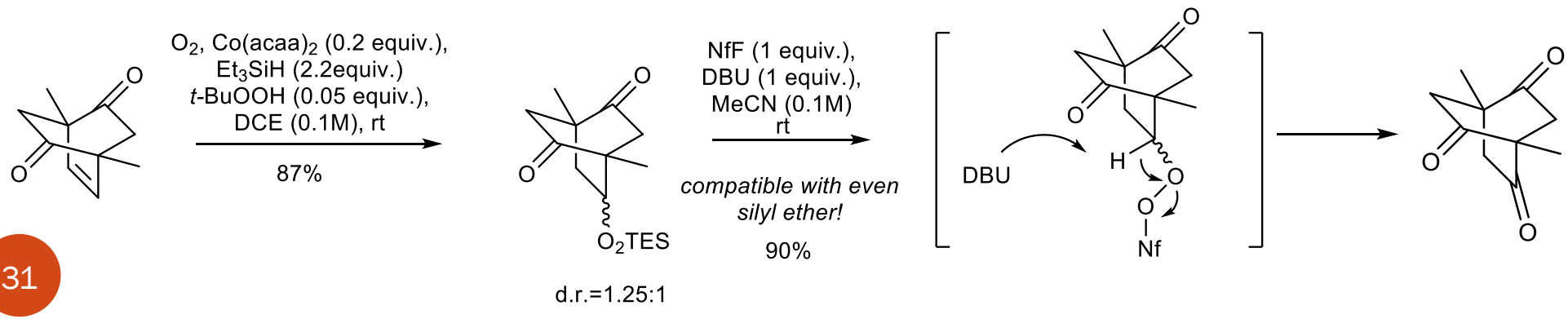
- The mechanism of this transformation has been studied and proposed by Nojimma et, al. in 2002. This is a radical-based reaction.



Org. Lett. **2002**, *4*, 3595-3598

Screening the Condition & Model System Study

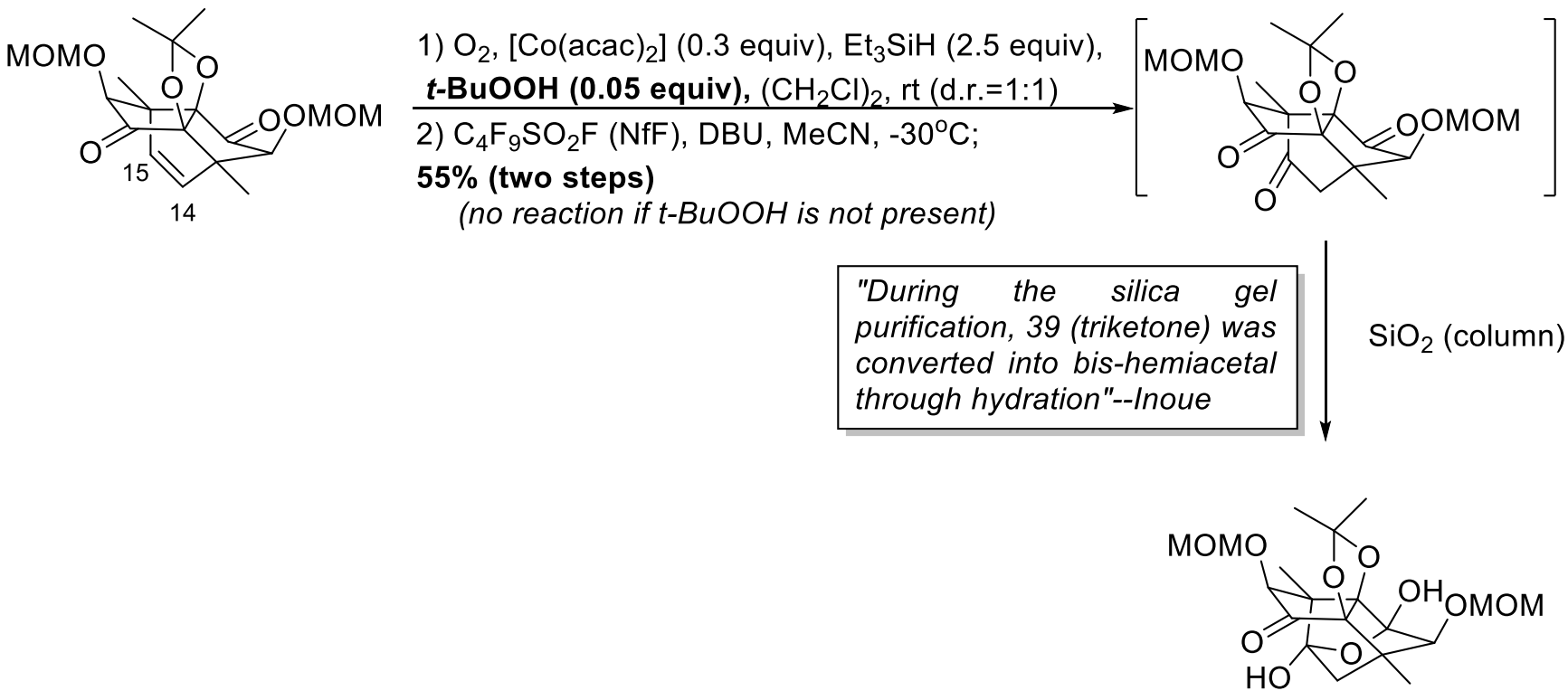
- Catalytic amount of *t*-BuOOH was found to accelerate hydroperoxysilylation (presumably by shortening the induction period)
- NfF ($C_4F_9SO_2F$) and DBU condition promotes the elimination of NfOH and ketone formation while being compatible with many functional groups including ketone and silyl ether. MsF and TsF are also found to be effective (all contain RSO_2F motif).



Bull. Chem. Soc. Jap. **1990**, *63*, 1305-1310

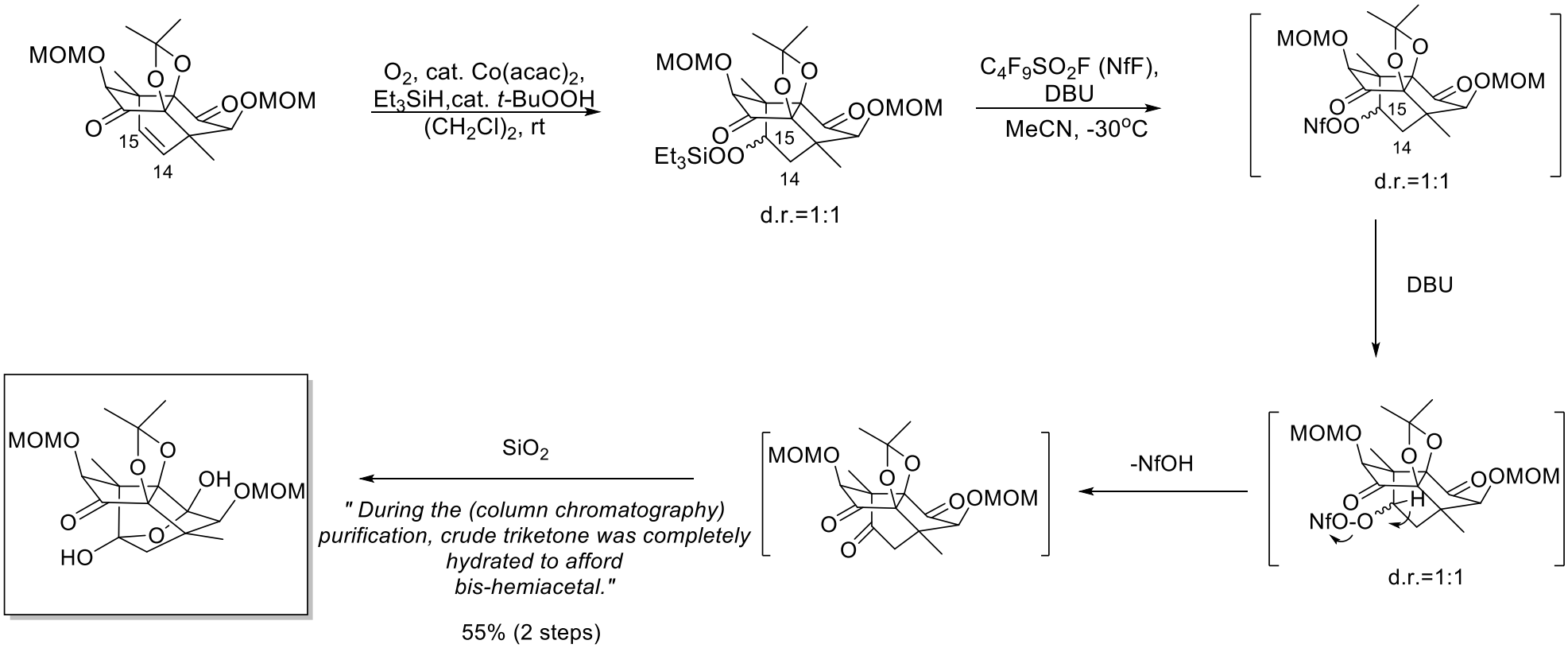
Application in Real System

- *t*-BuOOH is essential for Mukaiyama-Isayama Hydroperoxysilylation to work in real system.
- A note about chirality here: C15 and C14 are equivalent in C2 Symmetric tricycle. C2 Symmetry is destroyed after oxidation. However, only one enantiomer is obtained because C15 and C14 ***was*** equivalent.

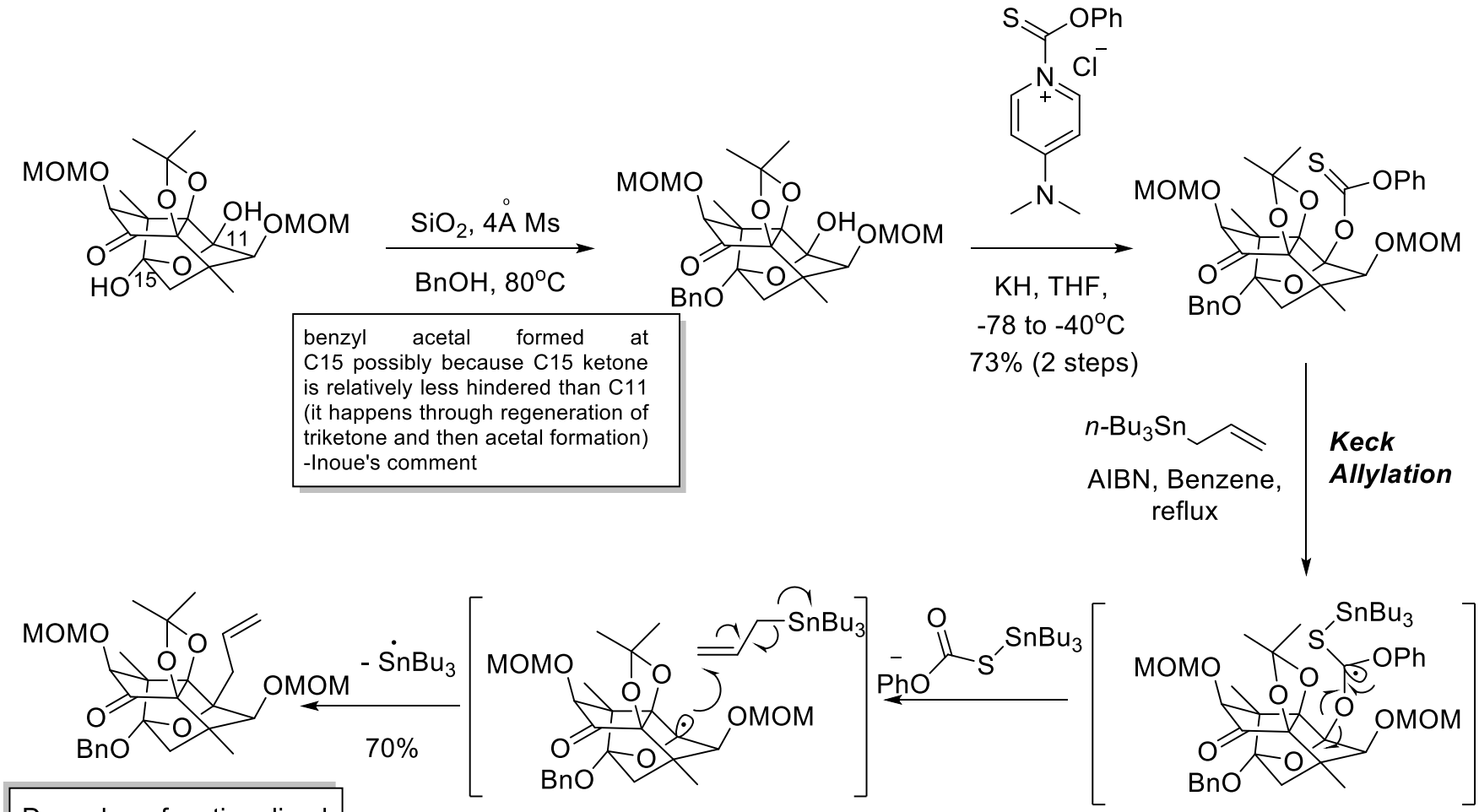


Synthesis of ABDE-ring

- Desymmetrization of C2-symmetric diketone



Construction of ABDE-ring: Final Stage



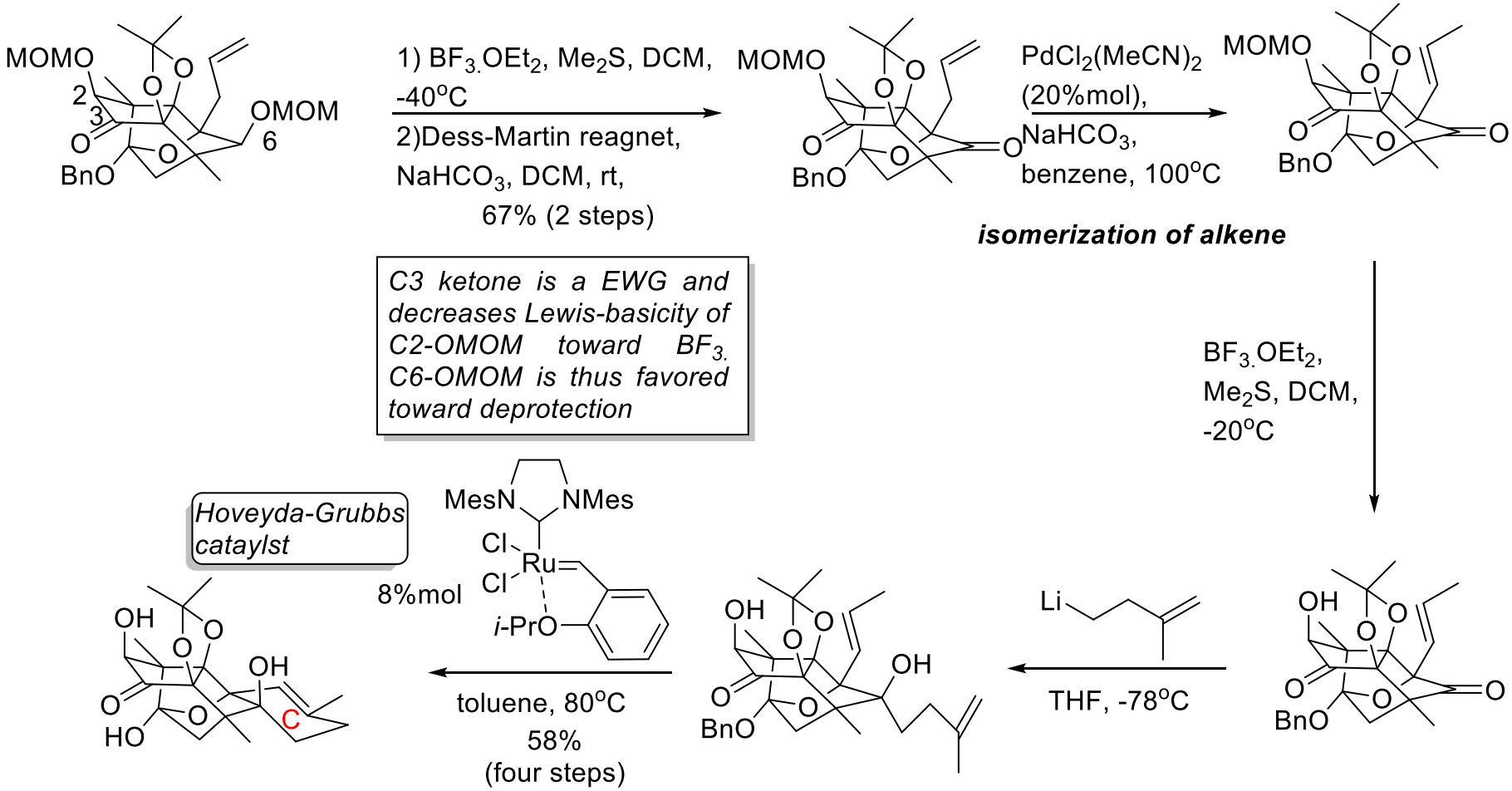
benzyl acetal formed at C15 possibly because C15 ketone is relatively less hindered than C11 (it happens through regeneration of triketone and then acetal formation) -Inoue's comment

α -alkoxy bridgehead radical

Densely functionalized ABDE ring was synthesized in 21 steps from 2,5-dimethylbenzene-1,4-diol

C-Ring Construction

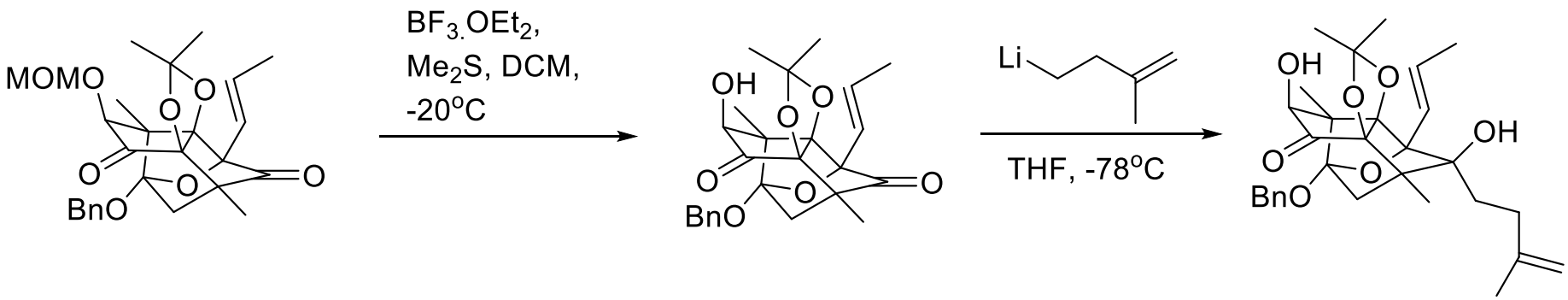
- Key strategy: RCM



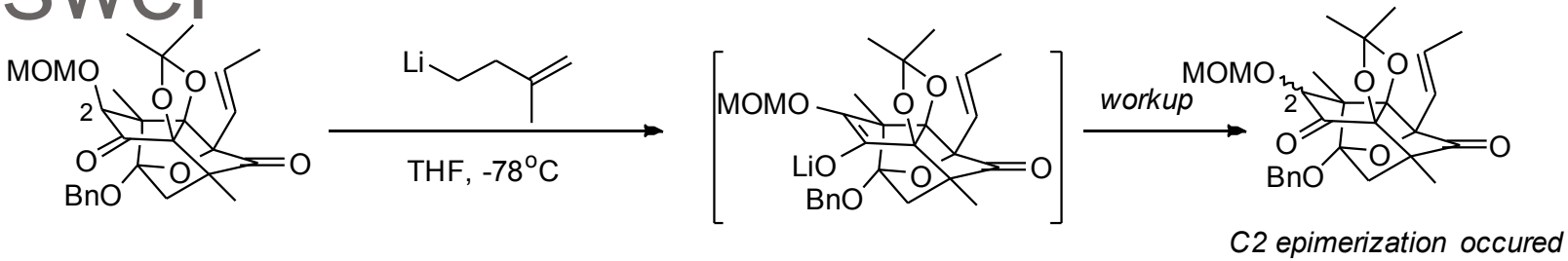
C3 ketone is a EWG and decreases Lewis-basicity of C2-OMOM toward BF_3. C6-OMOM is thus favored toward deprotection

Group Meeting Problem

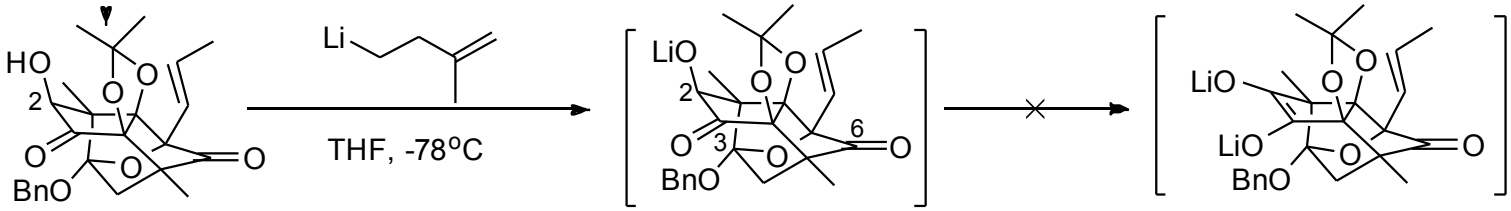
- Could you take a guess about why the author removed MOM protection first and then do the alkylation? Also, could you rationalize the regio&stereoselectivity of the alkylation step?



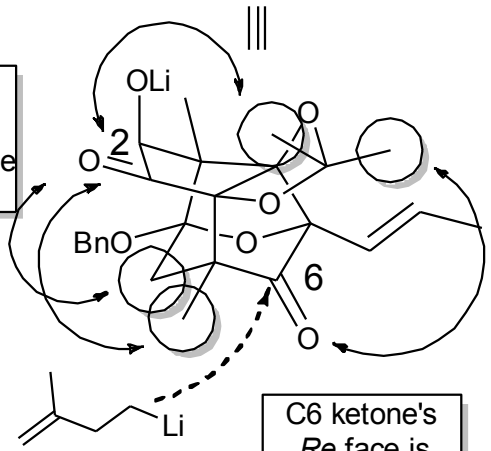
Answer



C2 epimerization occured



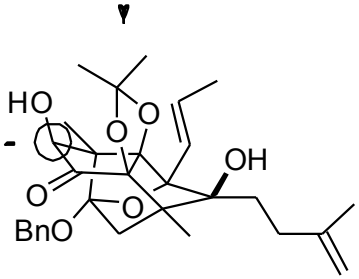
Dianion is way harder to form! epimerization is suppressed!



Methylene and methyls block both face of C2 ketone

Si face of C6 ketone is blocked by acetonide's methyl

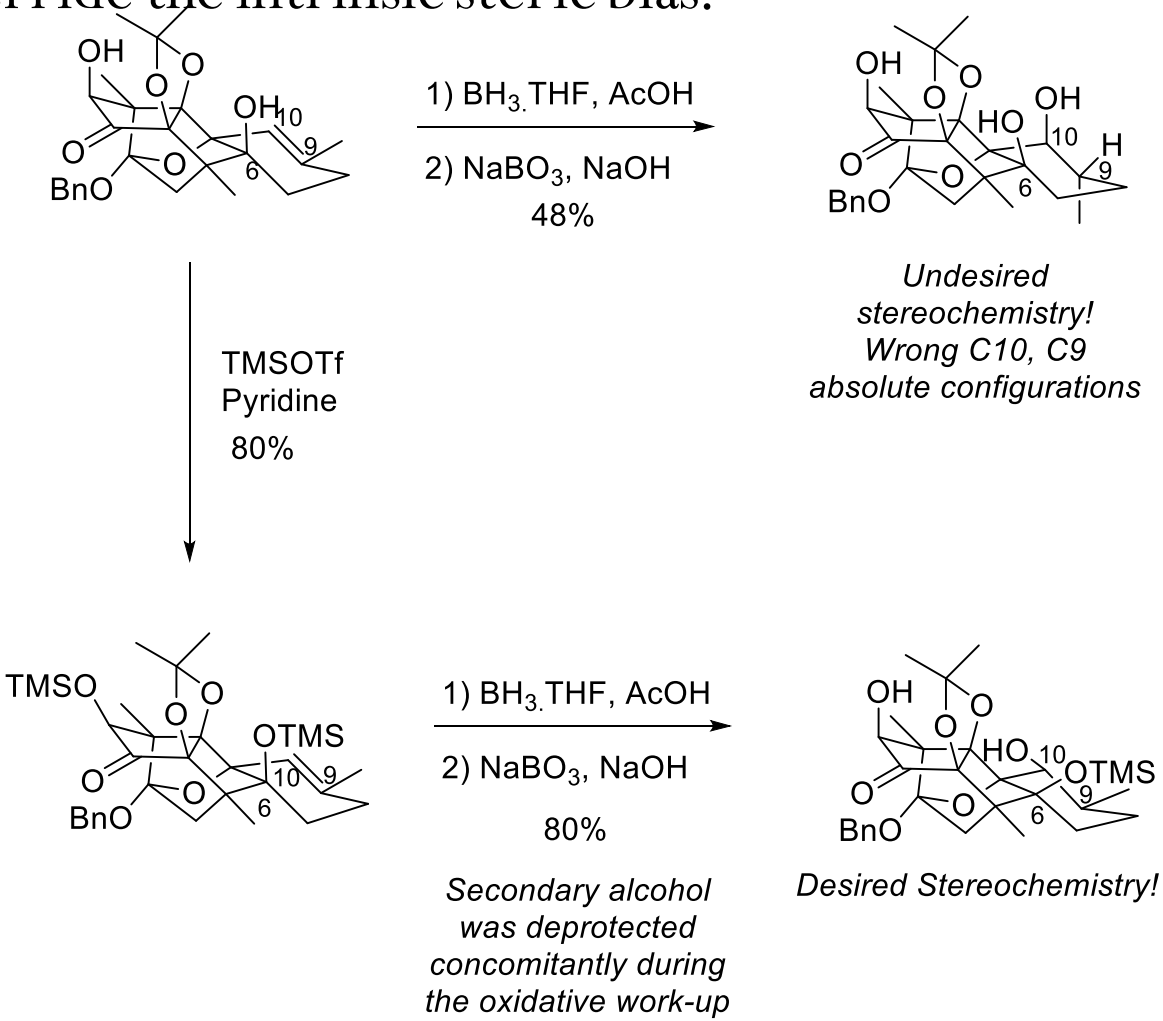
C6 ketone's *Re* face is relatively less hindered



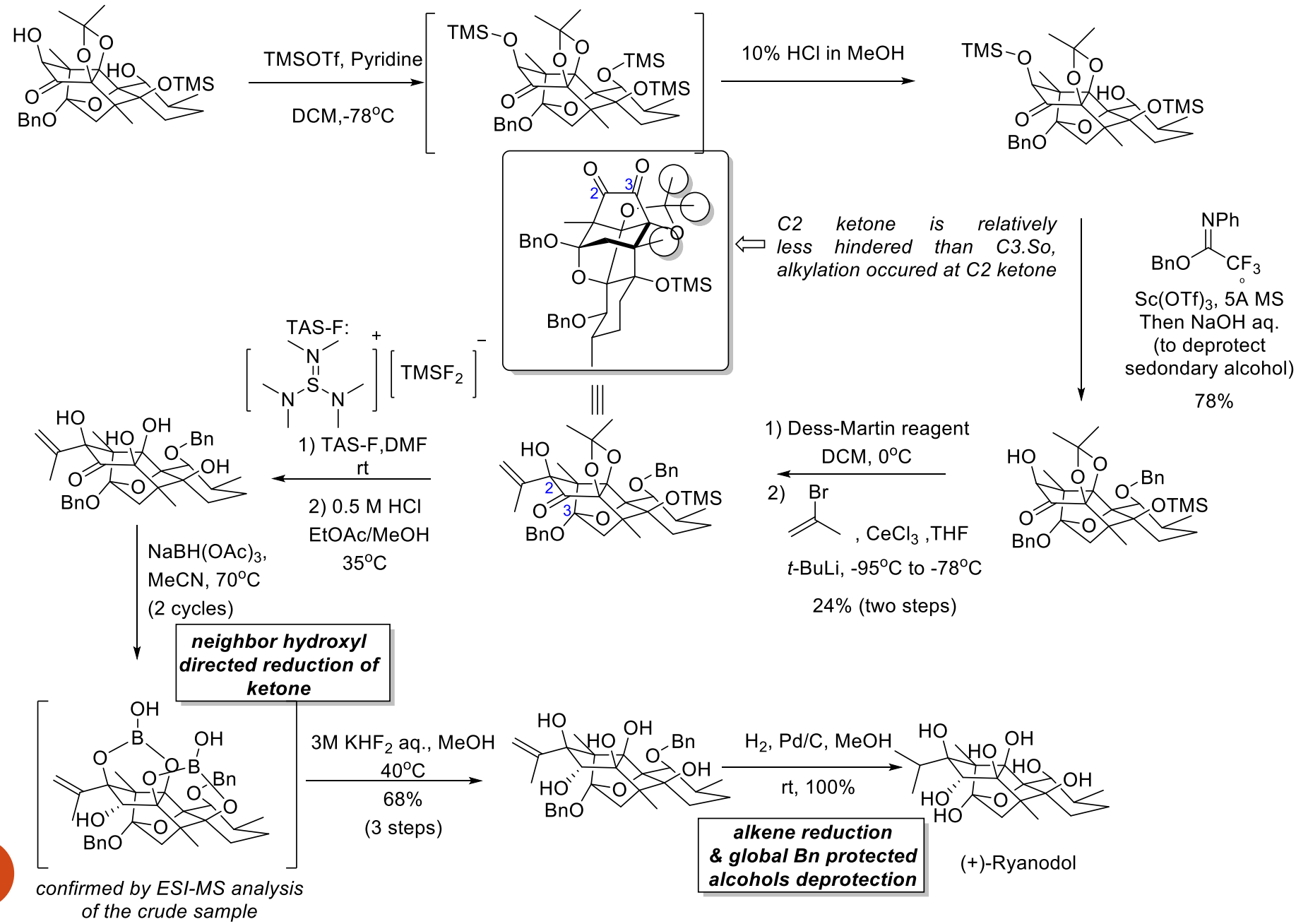
They tried to save this C2 stereocenter because it has an influence on the selectivity of later hydroboration step

C9,C10-stereocenters Construction

- Introduction of TMS on C6 hydroxyl is essential for the needed stereoselectivity of hydroboration/oxidation step to happen. TMS helps override the intrinsic steric bias.

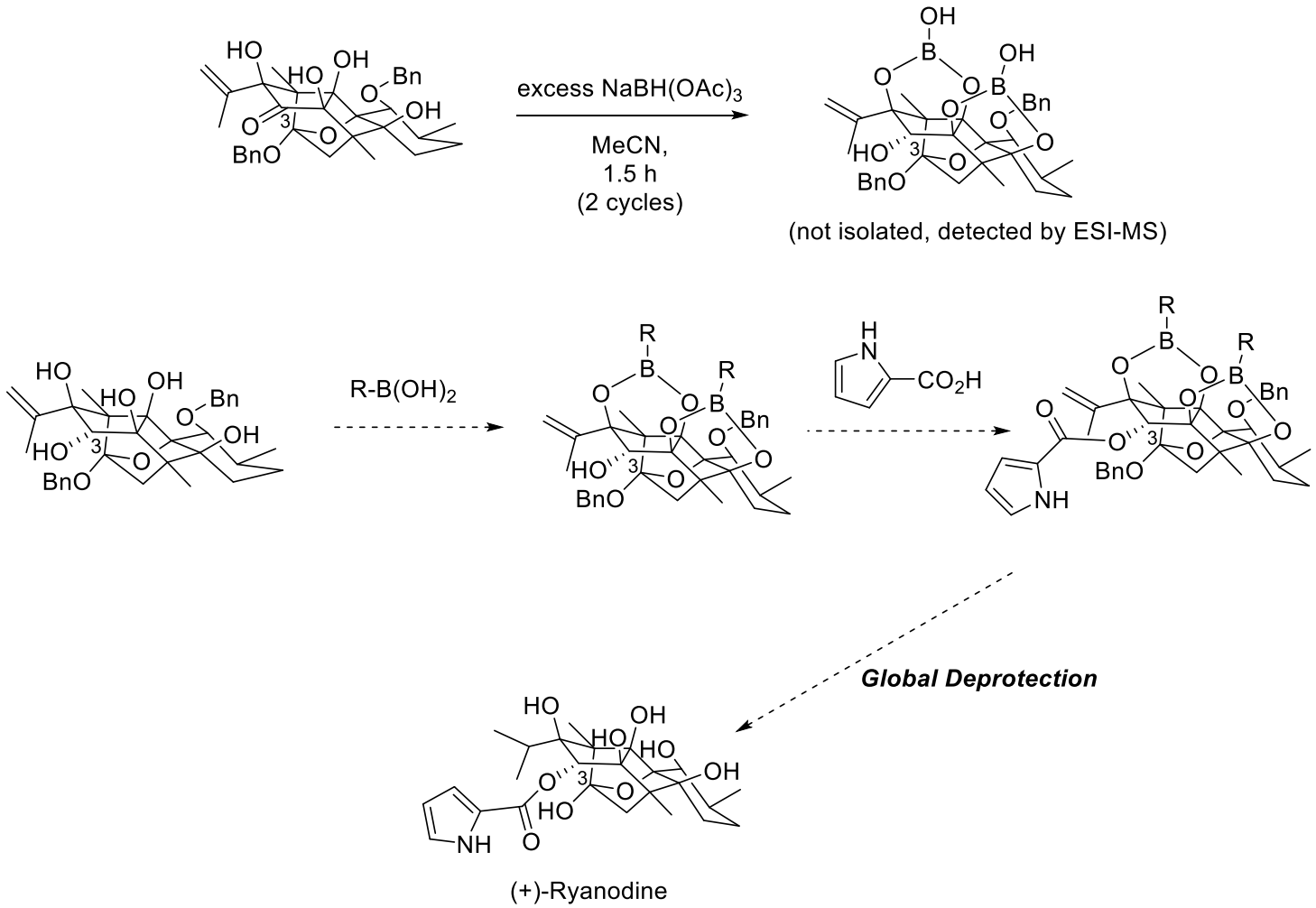


Endgame for (+)-Ryanodol



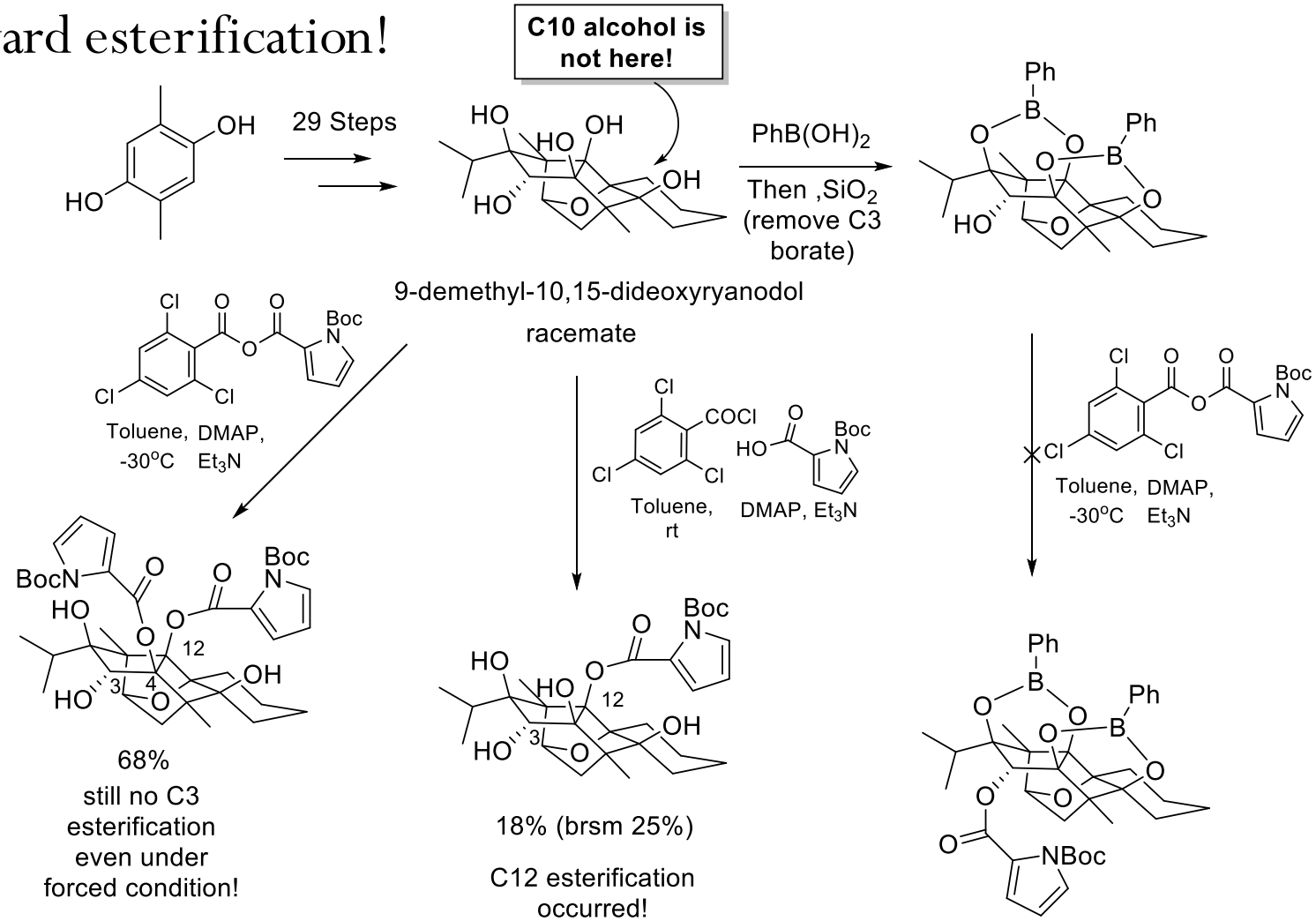
Toward (+)-Ryanodine: Protection Strategy Development

- From Deslongchamps' study, we knew that direct esterification on Ryanodol is not possible(C10 not C3 is favored position).
- Inspired by C3 ketone reduction during total synthesis of (+)-Ryanodol , Inoue group decided to use borate ester to protect all four tertiary alcohols in order to synthesize (+)-Ryanodine



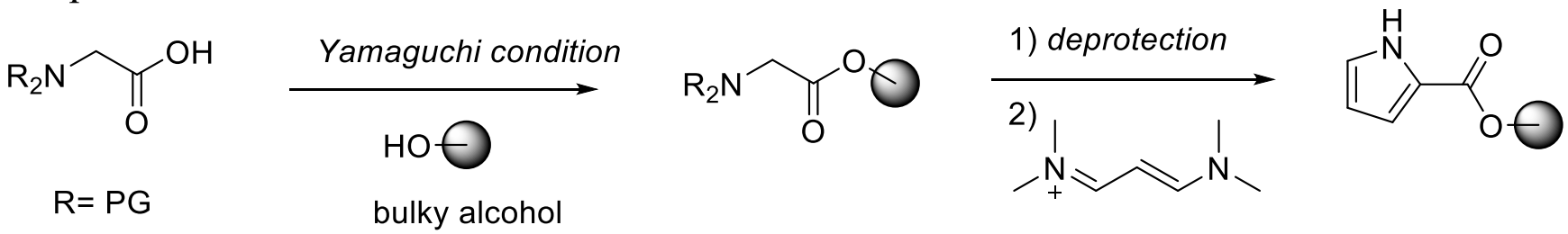
Model Study

- C3 alcohol is so hindered that, even under forced condition, there is no reaction!
- C3 alcohol has lower reactivity than tertiary alcohols in the system toward esterification!



New Strategy toward Pyrrole Ester Synthesis

- Direct introduction of pyrrole moiety is impossible. C3 alcohol is too hindered for this transformation.
- Hint: to have a faster esterification, one can use less hindered alcohol or/and acid.
- Inoue's strategy: Use "smaller" acid and then construct pyrrole ring in a 2-step manner.



R= PG

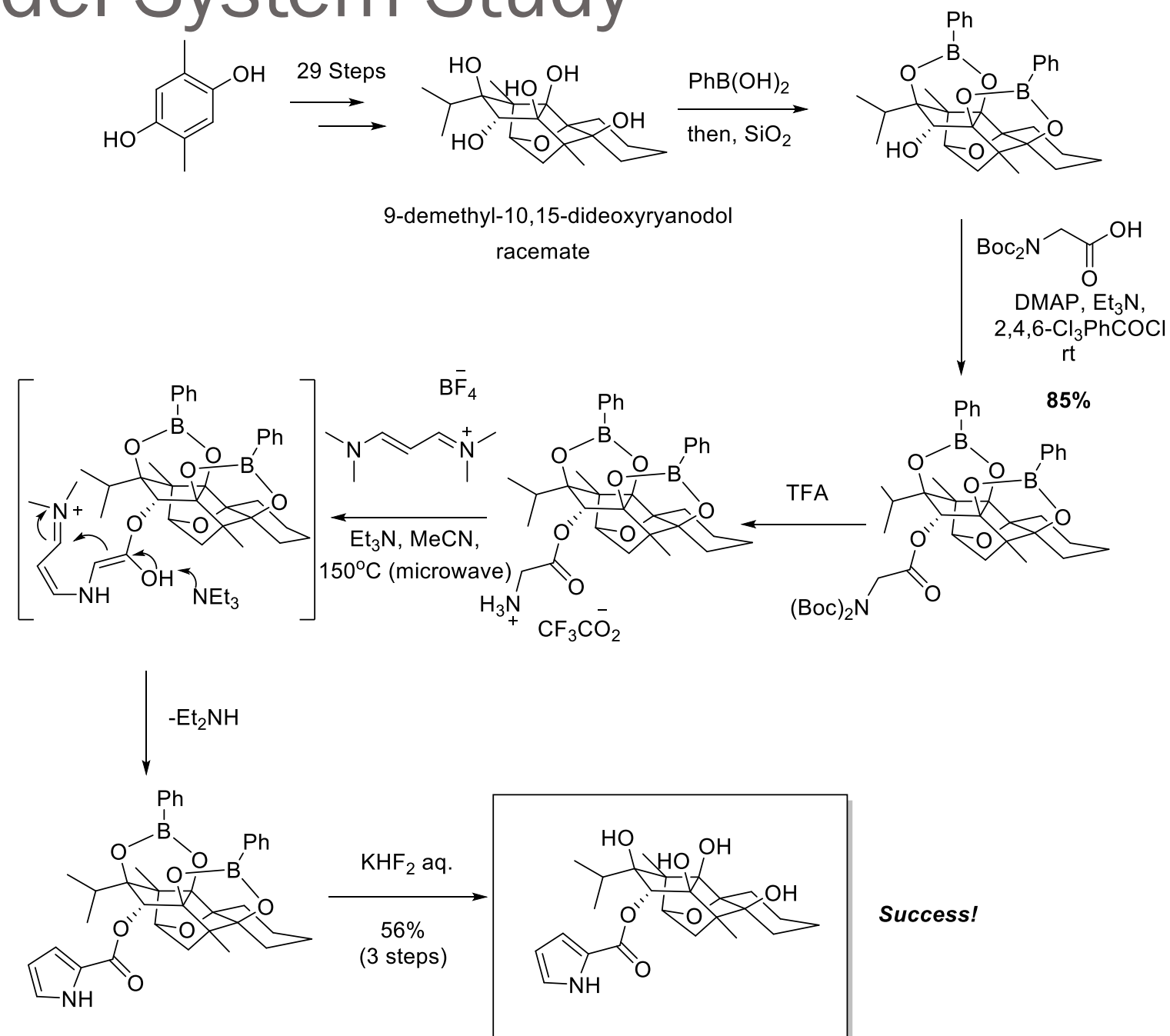
glycine derivative as *less* hindered, more electron-deficient pyrrole equivalent

bulky alcohol

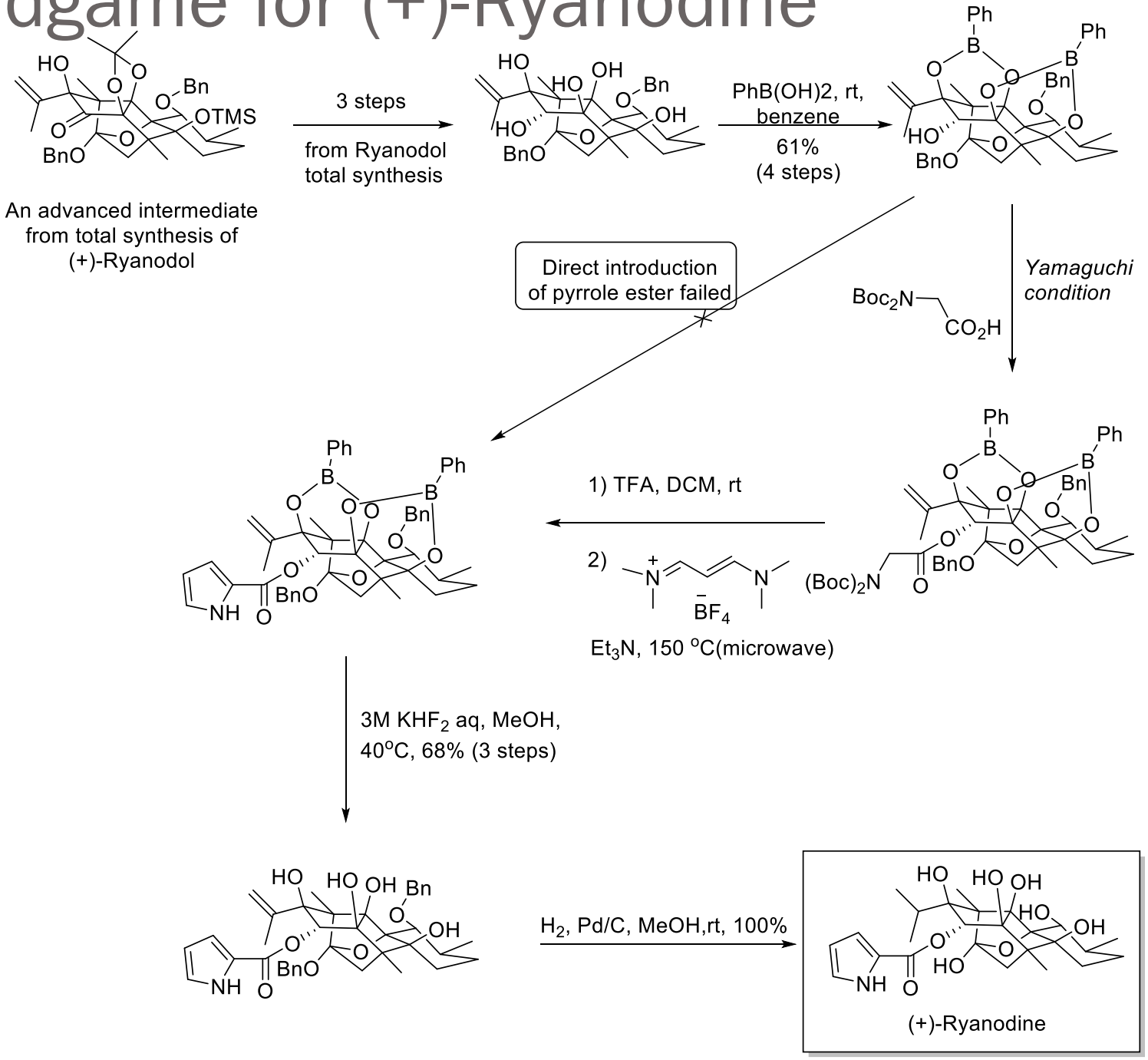
J. Org. Chem., **1990**, *55*, 4735-4740

Relatively electron deficient alkyl-substituent of amino acid may also increase the reactivity of the carbonyl of –COOH --- also facilitate esterification!

Model System Study



Endgame for (+)-Ryanodine



Summary

- First total synthesis of (+)-Ryanodine. Borate ester protection strategy and 2-step pyrrole ester synthesis from (+)-Ryanodol solved the 36-year-old puzzle.
- Applied symmetry-driven strategy in Ryanoid core synthesis, which increases the efficiency of the synthesis.
- 37 steps, linear sequence, toward (+)-Ryanodol total synthesis. 42 Steps, linear sequence, toward (+)-Ryanodine total synthesis.