Total Synthesis of
Some Bioactive Natural Products
Having Polycyclic Systems

連続多環式骨格を有する

数種の天然生理活性物質の全合成

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Research of Bioactive Substances Science

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Contents

Preface

A large number of bioactive natural products have ever been isolated and studied in various research fields for their chemical and medical applications. Total synthesis has been fundamental to those research fields and, especially, essential to the drug discovery. Therefore, total synthesis of natural products possessing a new class of framework and various analogues with their interesting bioactivities is worthwhile. In order to maximize benefit from total synthesis, total synthesis has to be as efficient and diverse to synthesize abundant and various analogues as possible.

Although synthetic methodologies have been developing, lots of problems have remained unsolved in total synthesis of natural products. Efficient and stereoselective synthesis of polycyclic ring systems has been one of those problems. Natural products and drugs often have their own chiralities, and their structures have a relationship to their bioactivities. Thus, it is extremely important to synthesize target materials stereoselectively and to determine the three-dimensional structure. Efficient synthesis of target materials accelerates the studies toward the drug discovery, and gives the diversity to the researches.

The title of this doctoral dissertation is total synthesis of some bioactive natural products having polycyclic systems. Two natural products described in this thesis, K1115B$_1$s and TMC-66, had been such problematic compounds that their absolute structures had never been determined and they had their own new polycyclic frameworks including stereogenic centers. We tried to resolve these problems by convergent and diverse total syntheses.

In the first chapter, the first total synthesis and structural determination of K1115B$_1$s (alnumycin) have been achieved. K1115B$_1$ has a sugar-like chiral 1,3-dioxane moiety attaching to pyranonaphthoquinone skeleton which belongs to a new class of pyranonaphthoquinone antibiotics, named naphthopyranomycin family, and only the relative stereochemistry of the partial structure of naphthopyranomycin family including K1115B$_1$ had been determined. And it had a difficulty in its structural determination owing to the distance between its chiral centers. Therefore, the convergent and stereoselective route from two sugars as the chiral resources has been established.

In the second chapter, the first total synthesis and structural determination of TMC-66 have been achieved in 9 steps as the longest linear sequence. TMC-66 has a benzo[*a*]naphthacenequinone skeleton fused with an oxazolidine ring and its absolute structure was not determined. Although benzo[*a*]naphthacenequinone antibiotics have ever been reported, a polycyclic structure with an oxazolidine is rare. And benzo[*a*]naphthacenequinone structure might be responsible for exhibiting endothelin converting enzyme inhibitory activity. As a result of numerous experiments, the regioselective intramolecular oxidative coupling was realized in high yield using Cu-NMI complex. The short-step and stereoselective route to the benzo[*a*]naphthacenequinone fused with an oxazolidine ring including stereogenic centers has been established.

Abbreviations

Ac	acetyl	Ph	phenyl
AcOH	acetic acid	PhH	benzene
aq.	aqueous	PhMe	toluene
Bn	benzyl	PIFA	[bis(trifluoroacetoxy)iodo(Ⅲ)]benzene
Bu	butyl	Pr	propyl
CAN	ceric(Ⅳ) ammonium nitrate	*p*-TsOH	*p*-toluenesulfonic acid
cat.	catalytic	Py	pyridine
CSA	10-camphorsulfonic acid	TBAF	tetra-*n*-butylammonium fluoride
dba	*trans*, *trans*-dibenzylidenacetone	TBAI	tetra-*n*-butylammonium iodide
DIBAL	diisobutylaluminium hydride	TBS	*tert*-butyldimethylsilyl
DMAP	4-dimethylaminopyridine	Tf	trifluoromethanesulfonyl
DMF	*N,N*-dimethylformamide	TMS	trimethylsilyl
DMSO	dimethylsulfoxide	THF	tetrahydrofuran
Et	ethyl	TMEDA	tetramethylethylenediamine
EtOAc	ethyl acetate	TBSOTf	*tert*-butyldimethylsilyl triflate
HMPA	hexamethylphosphoramide	TMSOTf	trimethylsilyl triflate
LDA	lithium diisopropylamide		
LHMDS	lithium bis(trimethylsilyl)amide		
*m*CPBA	*m*-choroperbenzoic acid		
Me	methyl		
MeCN	acetonitrile		
MOM	methoxymethyl		
Ms	mesyl		
NBS	*N*-bromosuccinimide		
NHMDS	sodium bis(trimethylsilyl)amide		
NMI	*N*-methylimidazole		

# Chapter 1. The First Total Synthesis and the Structural Determination of Antitumors K1115 B_1s (alnumycins).

Section 1. Introduction

Naphthopyranomycin family, involving naphthopyranomycin (**1.1**)[1], K1115 B_1 (**1.2**)[2], and exfoliamycin (**1.3**)[3] (Figure 1.1) has been studied as a unique group of bioactive natural products. Naphthopyranomycin family possesses various bioactivities, for example, antibacterial activity, antitumor activity, IL-1β converting enzyme inhibitory activity, and so on. Structurally, naphthopyranomycin family has a chiral sugar-like ring attaching to pyranonaphthoquinone framework. This sugar-like 1,3-dioxane moiety is uncommon in natural products, and there are only a little knowledge about its biosynthesis.[4] Although naphthopyranomycin family has been isolated widespread, their absolute structures have never been disclosed. In our laboratory, the total synthesis of bioactive pyranonaphthoquinones including nanaomycin[5], kalafungin[5], medermycin[6], BE-54238B[7], and BE-52440A[8] have been achieved (Figure 1.2). The structure and bioactivities of naphthopyranomycin family drew our attention to determine the absolute structure.

K1115B_1 (**1.2**) was a member of naphthopyranomycin family, which was isolated independently from *Streptomyces* sp. A41956 as BE-41956A (Banyu group, 1997)[9], *Streptomyces* sp. DSM 11575 as alnumycin (Bieber et al., 1998)[10] and *Streptomyces griseorubiginosus* strain Mer-K1115 as K1115 B_1 (Eisai and Mercian group, 1998)[2]. K1115 B_1 (**1.2**) shows several biological activities, including antibiotic, cytostatic, gyrase inhibitory, and topoisomerase inhibitory activity.[11] Structurally, K1115B_1 (**1.2**) has three features compared with the other members of the naphthopyranomycin family: 1) 1,3-dioxane moiety, 2) the chiral carbon at C3 position missing by the double bond formation between C3 and C4, and 3) no existence of a side chain at C7 position. And even its partial relative stereochemistry had never been determined due to the distance between chiral centers. Herein, we report the first total synthesis and the structural determination of K1115 B_1 (**1.2**).
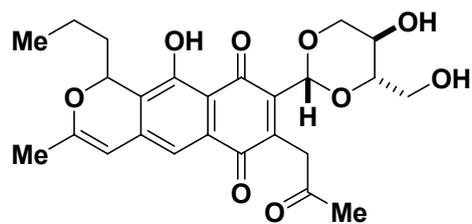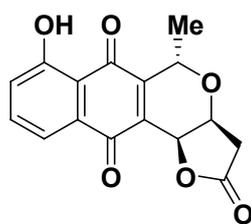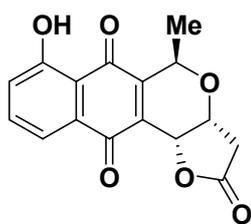
Naphthopyranomycin (1.1)
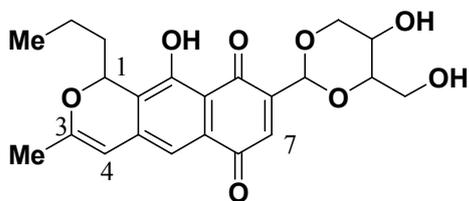
K1115B$_1$ (1.2)
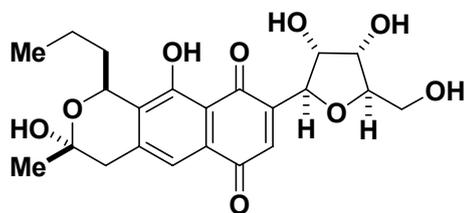
Exfoliamycin (1.3)

Figure 1.1. Examples of naphthopyranomycin family and structure of K1115 B$_1$.

Nanaomycin

Kalafungin
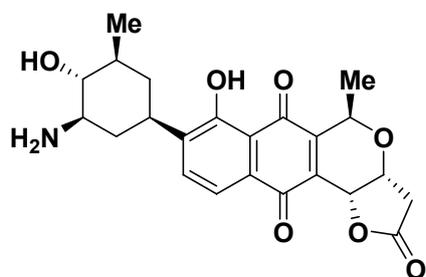
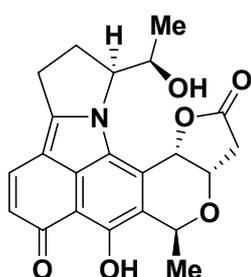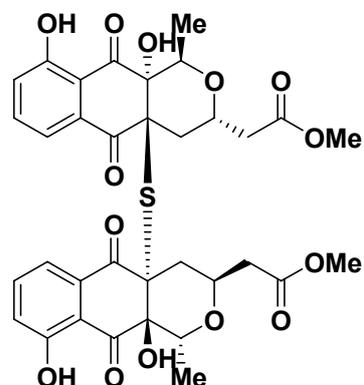**Medermycin**

BE-54238B

BE-52440A

Figure 1.2. Natural products possessing a pyranonaphthoquinone skeleton.

Section 2. Retrosynthetic Analysis

In order to determine the absolute structure of K1115 B₁, its four diastereomers would be synthesized stereoselectively (Figure 1.3).

There were two challenges to synthesize K1115 B₁: 1) how to construct its chiral centers located at the both sides of the molecule. 2) how to construct its new pyranonaphthoquinone skeleton. In cases that various analogues of naphthopyranomycin family were synthesized, these two factors are very important, too. Especially, it is exclusively essential to establish a convergent and efficient route to connect the main framework and the acetal unit. Therefore, in our plan, stereogenic centers at the both sides would be constructed respectively using two kinds of sugars as each starting material (Scheme 1.1).

K1115 B₁ would be synthesized from the compound **1.4** by acetal formation with diol **1.5** or ***ent*-1.5** provided from D-glucose. And stereoselective propylation of **1.6**, controlled on the basis of its own chiralities, would afford **1.4**. Tricyclic **1.6** would be synthesized by coupling between **1.7** and **1.8** by Michael-Dieckmann type annulation. Lactone **1.7** has been synthesized in 3 steps from 3,4-di-*O*-acetyl-6-deoxy-L-glucal in the previous paper[12].
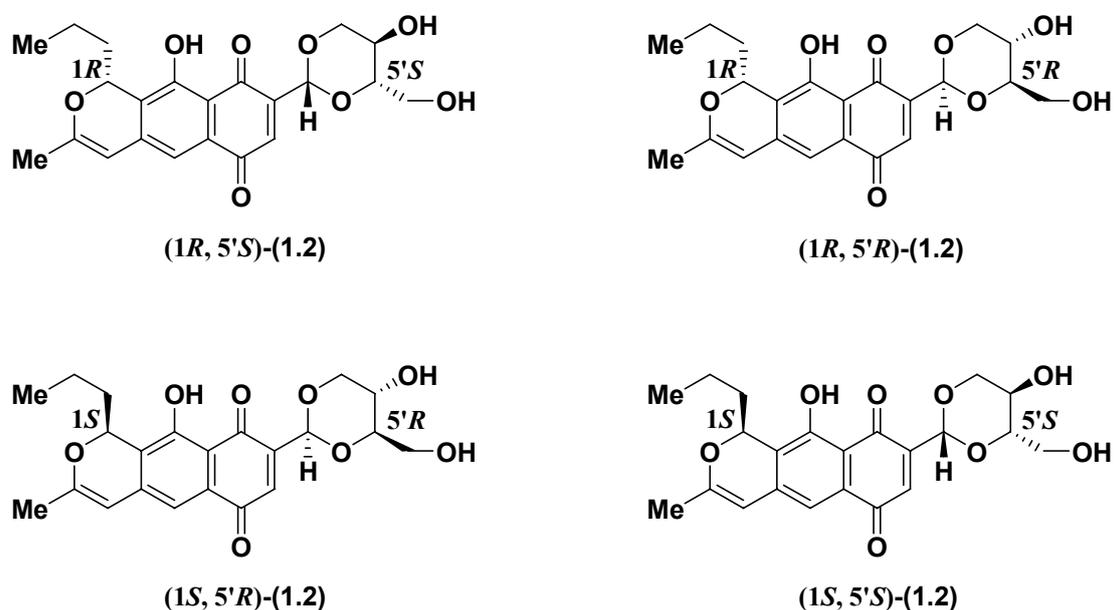

Figure 1.3. Four diastereomers of K1115 B₁.
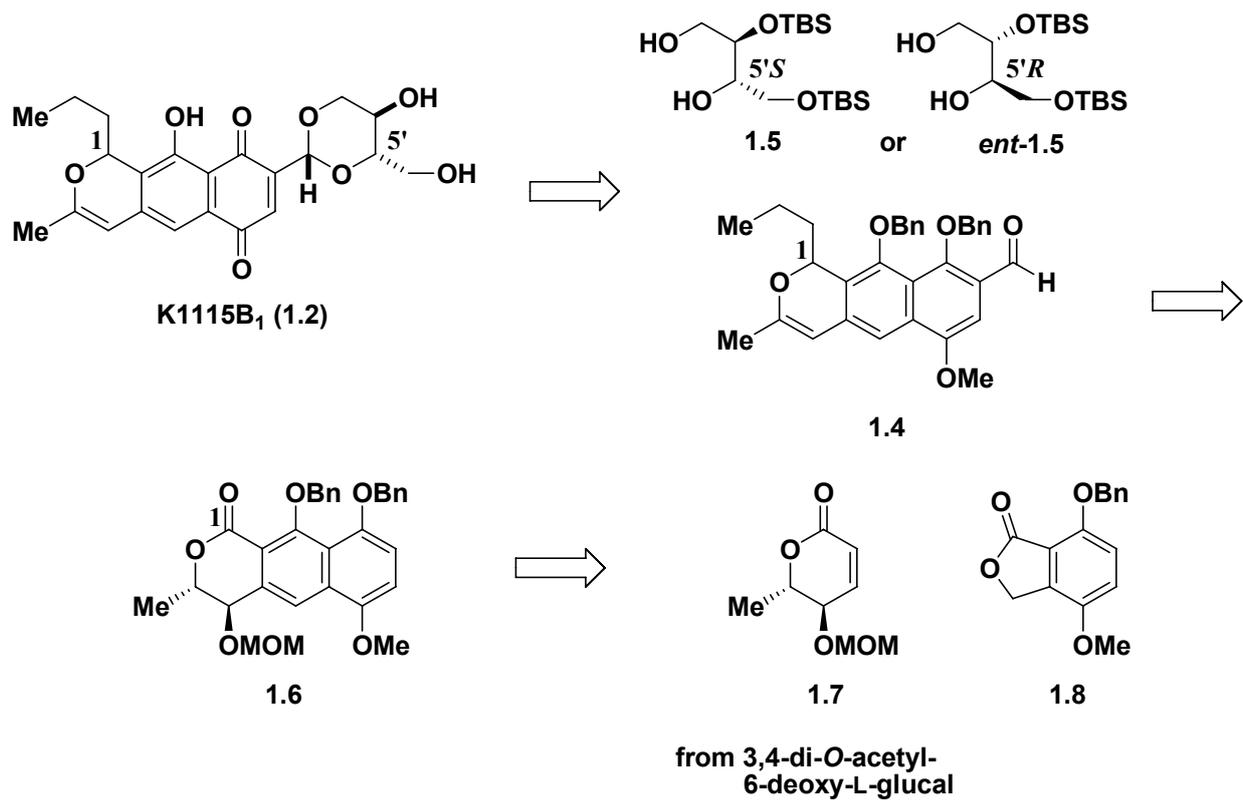
Scheme 1.1. Retrosynthetic analysis of K1115 B₁.

Section 3. Total Synthesis and Structural Determination of the Target Materials

Part 1. Syntheses of the Michael Donor and Michael Acceptor, and Tandem Michael-Dieckmann Type Annulation.

Michael donors **1.11** and **1.8** were synthesized in 2 and 4 steps, respectively (Scheme 1.2). Reduction of 2,5-dimethoxybenzaldehyde with $LiAlH_4$ proceeded smoothly to give the corresponding benzyl alcohol **1.10** in 92% yield, which was followed by lactone formation to provide the Michael donor **1.11**[13]. Furthermore, exchange of the protective group of the hydroxyl group at the *ortho*-position of carbonyl group from methyl to benzyl group gave the other Michael donor **1.8**.
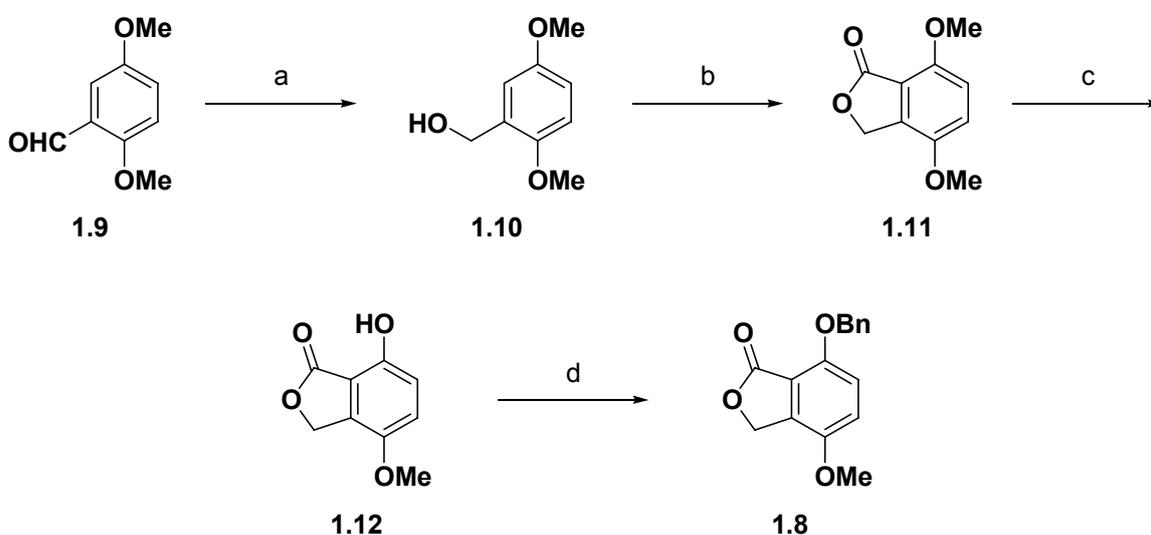
Scheme 1.2. Synthesis of the Michael donors **1.11** and **1.8**. Reagents and conditions: (a) $LiAlH_4$, THF, 0 °C, 20 min, 92 %; (b) *n*-BuLi, THF, –78 °C to reflux, 3 h, then CO_2, –78 °C to rt, 1 h, then 2N HCl, rt, 3 h, 59 %; (c) $MgBr_2$, NaI, MeCN, reflux, 4 h; (d) BnBr, K_2CO_3, 18-crown-6, DMF, rt, 4 h 60 % (2 steps).

Michael acceptor **1.7** was synthesized in 3 steps from 3,4-di-*O*-acetyl-6-deoxy-L-glucal (**1.13**)[12] (Scheme 1.3). Treatment of the starting material with *m*CPBA and boron trifluoride diethyl ether complex gave lactone **1.14**. And deacetylation and the following methoxymethylation afforded the Michael acceptor **1.7**.
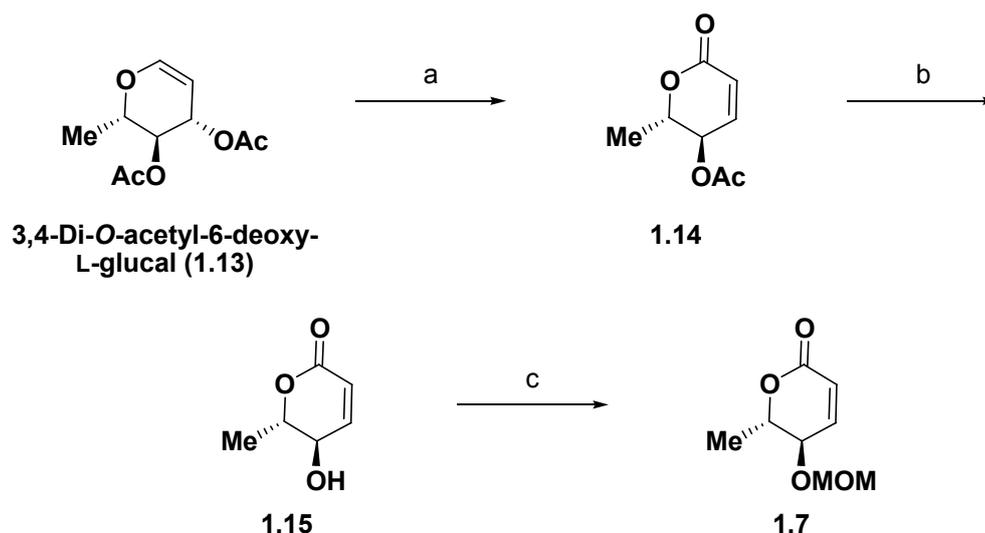
Scheme 1.3. Synthesis of the Michael acceptor **1.7**. Reagents and conditions: (a) BF$_3$ · OEt$_2$, mCPBA, CH$_2$Cl$_2$, −20 °C, 0 min., 79 %; (b) Lipase, phosphate buffer, rt, 2 d, 81 %; (c) MOMCl, i-Pr$_2$NEt, ClCH$_2$CH$_2$Cl, 70 °C, 2 h, 87 %.

Tandem Michael-Dieckmann reaction followed by aromatization proceeded to give **1.16** in 55% yield in 2 steps (Scheme 1.4). And benzyl protection afforded the tricyclic compound **1.17**.
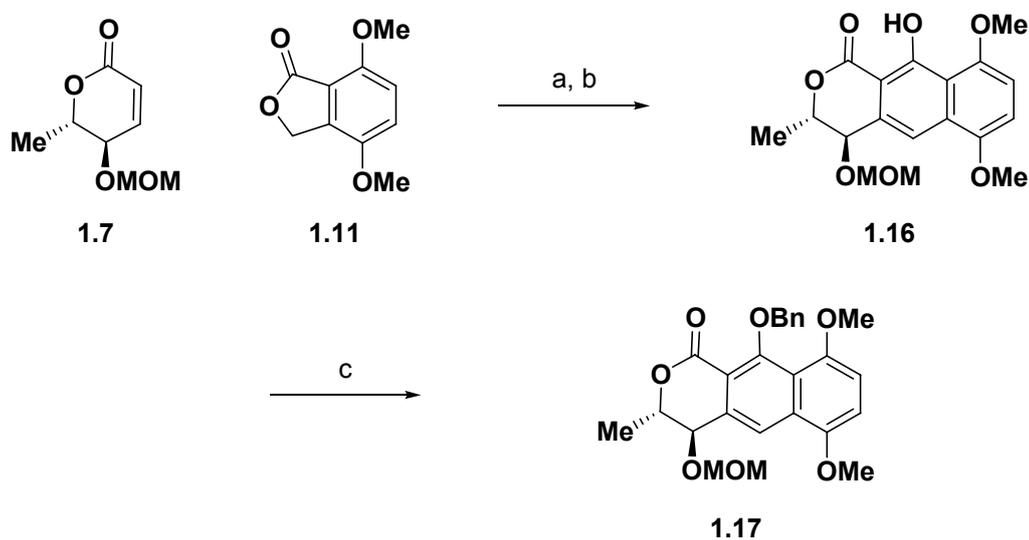
Scheme 1.4. Construction of tricyclic ring system by tandem Michael-Dieckmann. Reagents and conditions: (a) LHMDS, THF, −78 °C to rt, 1 h; (b) SOCl$_2$, Py, 0 °C, 0.5 h, 55 % (2 steps); (c) BnBr, K$_2$CO$_3$, 18-crown-6, acetone, 60 °C, 1 h, quant.

Part 2. Stereoselective Introduction of Propyl Unit and Regioselective Introduction of Formyl Group.

On the way to the total synthesis of K1115 B$_1$, stereoselective alkylation at the C1 position is crucial. It was proper to construct (1R)- or (1S)- configuration according to the selected conditions, enantiodivergently. As a result of various investigations, stereoselective introduction of propyl unit was accomplished as shown in Scheme 1.5.

Tricyclic lactone **1.17** was reduced with DIBAL to give the corresponding lactol, which was followed by the stereoselective allylation using allyltrimethylsilane and TMSOTf. These two reactions progressed without any purification by column chromatography since the exposure of the lactol to silica gel caused its own decomposition. The hydrogenation of *exo*-olefin gave the alkylated product **1.19** possessing (1S)-configuration.

Meanwhile, the (1R)-alkylated product **1.21** was provided as follows. Treatment of **1.17** with *n*-propylmagnesium bromide produced lactol **1.20**. And stereoselective hydrogenation of the cationic oxonium intermediate gave the alkylated product **1.21** possessing (1R)-configuration.

Their stereochemistry were determined by nOe experiments (ref. Part 5). Each alkylation mechanism was considered to be in a similar fashion as shown in Figure 1.4.
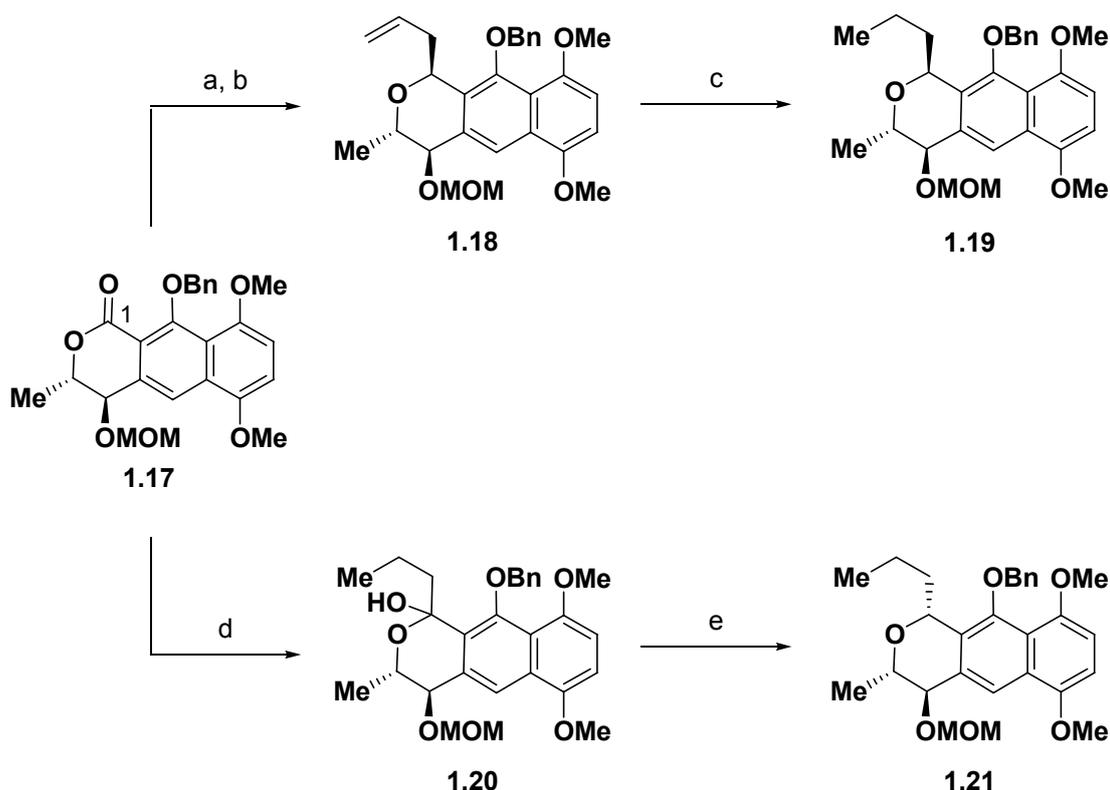
Scheme 1.5. Stereoselective introduction of propyl unit. Reagents and conditions: (a) DIBAL, PhMe, −78 °C, 2 h; (b) allyltrimethylsilane, TMSOTf, CH$_2$Cl$_2$, −78 °C, 5 min., 78 % (2 steps), (c) H$_2$, Pd/C, EtOAc, rt, 5 min., quant., (d) *n*-PrMgBr, THF, −78 °C, (e) H$_2$, Pd/C, EtOAc, 10 min., 60 %.
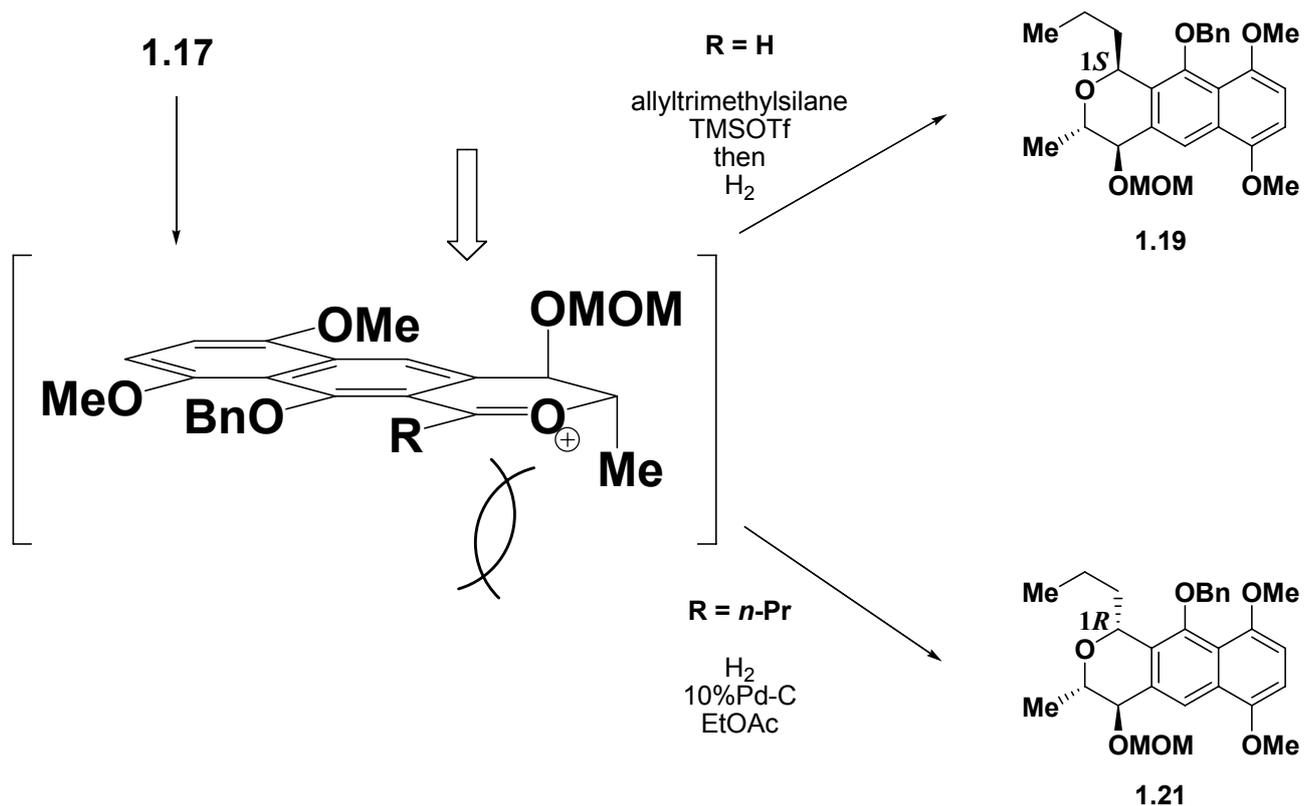
Figure 1.4. Possible mechanism of stereoselective propylation.

Moreover, Friedel-Crafts reaction of **1.19** gave the aldehyde **1.22** regioselectively as a single isomer, accompanying with deprotection of methoxymethyl group (Scheme 1.6). And dehydration with Burgess reagent afforded the compound **1.23**.
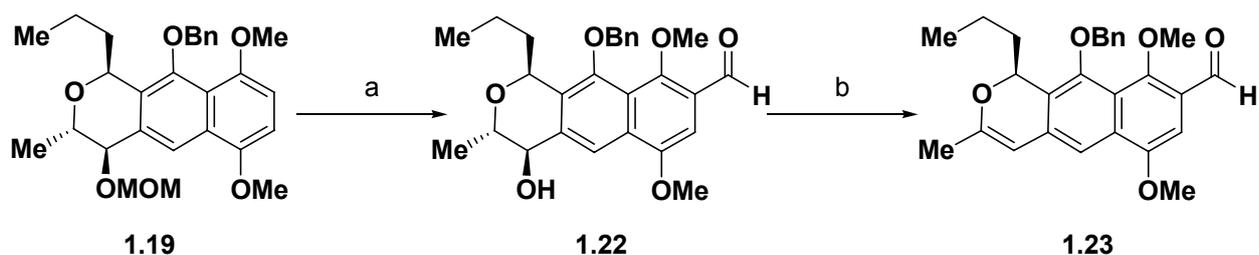
Scheme 1.6. Regioselective introduction of formyl unit. Reagents and conditions: (a) Cl_2CHOCH_3, $SnCl_4$, CH_2Cl_2, –78 °C, 8 min., 48 %; (b) Bergess salt, THF, 50 °C, 0.5 h.

Part 3. Syntheses of the Chiral 1,3-Butane-diols from D-Glucose for Constructing 1,3-Dioxane Moiety.

Protection of D-glucose by benzylidene acetal gave the compound **1.24** (Scheme 1.7). Oxidative cleavage and subsequent reduction provided diol **1.25**. Benzyl-protection and acidic hydrolysis afforded the benzyl-protected (5'*S*)-1,3-butane-diol **1.26**. In a similar way, TBS protected (5'*S*)-1,3-butane-diol **1.27** was obtained by TBS protection of **1.25** and hydrogenation. Furthermore, the benzyl-protected (5'*S*)-1,3-butane-diol **1.26** was converted into the TBS-protected (5'*R*)-1,3-butane-diol ***ent*-1.27** by TBS protection and debenzylation.
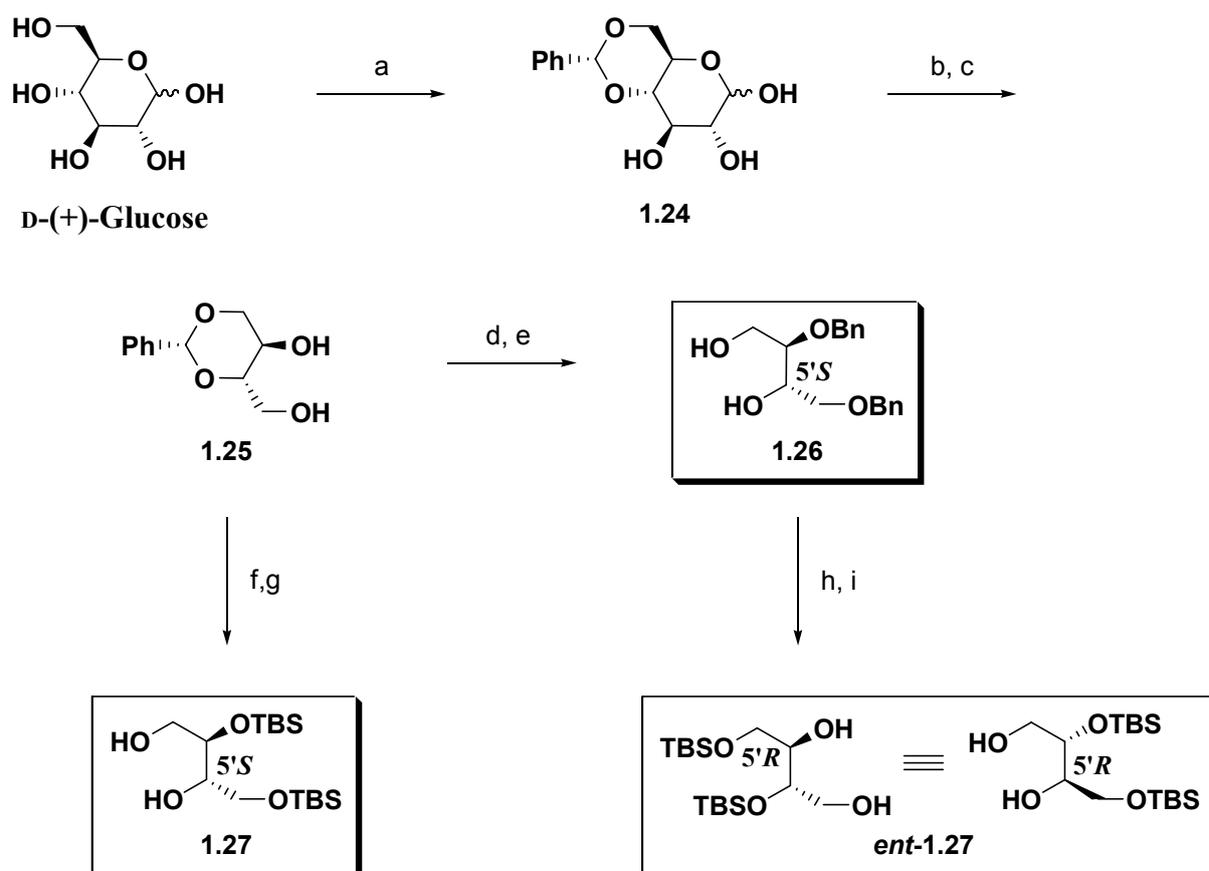
Scheme 1.7. Syntheses of the chiral 1,3-butane-diols. Reagents and conditions: (a) PhCH(OMe)$_2$, *p*-TsOH · H$_2$O, DMF, 30 mmHg, 60 °C, 2 h, 65 %; (b) NaIO$_4$, aq. EtOH, 0 °C, 1.5 h; (c) NaBH$_4$, MeOH, 0 °C, 1 h, 77 % (2 steps); (d) BnBr, NaH, DMF, rt, 1 h, 80 %; (e) 2N HCl, MeOH, reflux, 45 min., 93 %; (f) TBSOTf, 2,6-lutidine, CH$_2$Cl$_2$, rt, 2 h, quant.; (g) H$_2$, Pd(OH)$_2$, EtOH, rt, 40 min., 83 %; (h) TBSOTf, 2,6-lutidine, CH$_2$Cl$_2$, rt, 2 h, 94 %; (i) H$_2$, Pd(OH)$_2$, EtOH, rt, 1.5 h, 85 %.

Part 4. Stereoselective Construction of 1,3-Dioxane Moiety and Studies toward the Total Synthesis of the Target Materials.

Aldehyde **1.22** and diol **1.26** in hand, the stereoselective construction of 1,3-dioxane moiety was attempted (Scheme 1.8). There were two modes in the cyclization reaction, path A and B. The acetalization started from the attack of primary alcohol to the aldehyde of the compound **1.22**, followed by dehydration and formation of the oxonium cation intermediate **1.28**. Undesired cyclized product **1.29** was provided through path A in which the alcohol of intermediate **1.28** approached from the *Si* face of the oxonium carbonyl group. And path B explained that the attack of the alcohol from the *Re* face produced the desired product **1.30**. Path B was prior to path A since the desired product **1.30** was more stable than the undesired product **1.29** by 1,3-diaxial interaction. In this way, stereoselective construction of 1,3-dioxane moiety proceeded smoothly in 78 % yield to give a single diastereomer **1.30**.
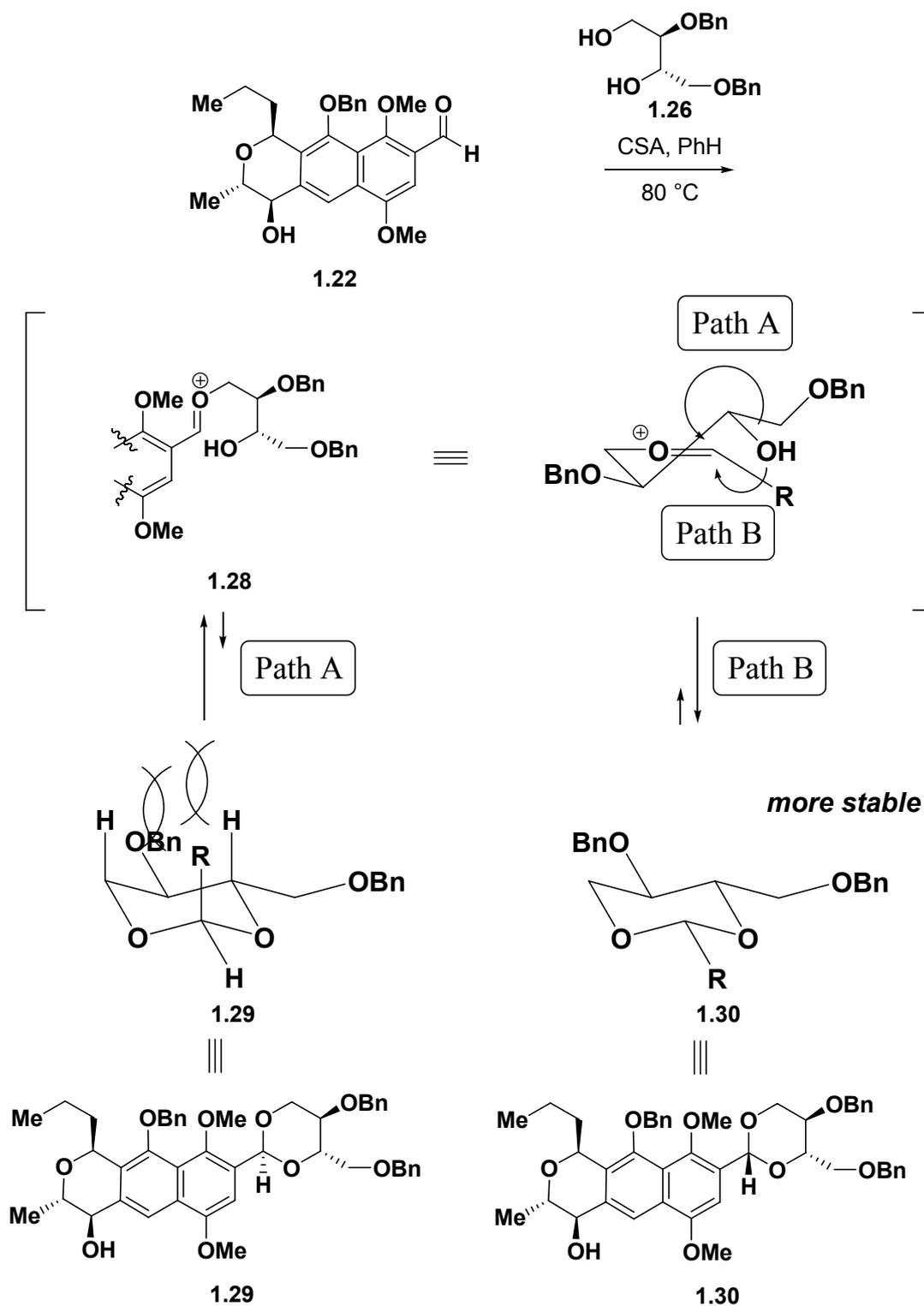
Scheme 1.8. Stereoselective construction of 1,3-dioxane moiety.

Further transformations toward K1115 B$_1$ were attempted. The cyclized product **1.30** was oxidized in@::to the naphthoquinone **1.31** (Scheme 1.9). Dehydration reaction, however, did not proceed. This dehydration reaction was attempted by conversion of the hydroxyl group into leaving groups, which found that the naphthoquinone **1.31** was decomposed under various conditions. For example, strong basic conditions such as NaH, NaH-DMSO and more weak basic conditions such as triethylamine gave some unidentified side products, and Mitsunobu conditions produced a little amount of the corresponding oxidized product. It was impossible to improve the stability of the naphthoquinone **1.31**. Thus, the regioselective deprotection of the phenolic benzyl group was investigated to react the hydroxyl group at the benzyl position with dehydrating reagents before decomposing itself. However, all attempts to the selective deprotection did not proceed efficiently, and only trace amount of **1.33** was obtained.
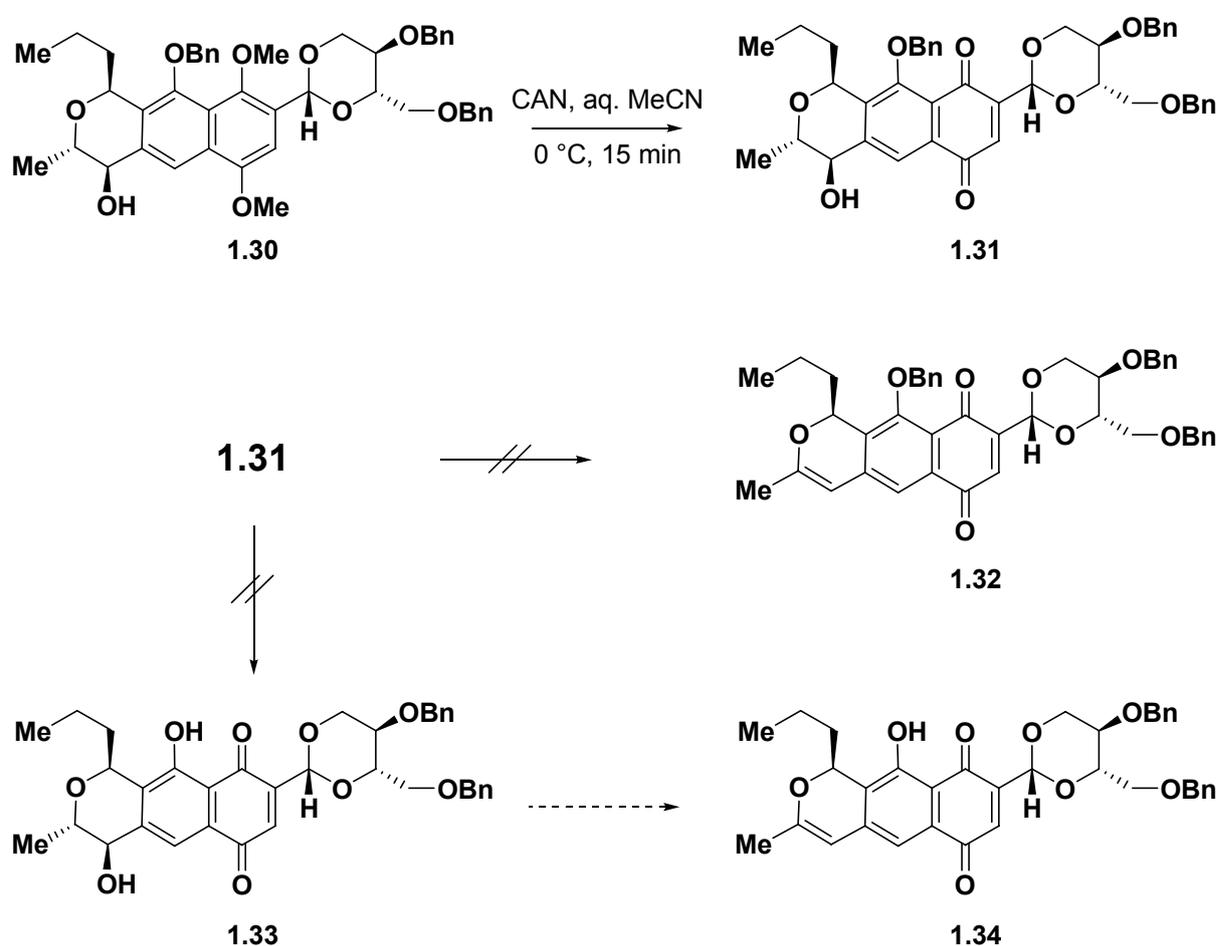
Scheme 1.9. Oxidation to *p*-Quinone and Investigations of Construction of K1115 B$_1$ Skeleton.

It appeared that the naphthoquinone **1.31** could not be converted into the K1115 B₁ as the results mentioned above. Thus, the K1115 B₁ skeleton was tried to construct via the dehydrated **1.23** (Scheme 1.6). Formation of the acetal group from the compound **1.23** using the benzyl-protected 1,3-butane-diol **1.26** afforded **1.35** possessing the 1,3-dioxane moiety (Scheme 1.10). Oxidation into the quinone **1.36**, however, gave a complex mixture, and decomposed the acetal unit before formation of the desired *p*-quinone **1.36**.
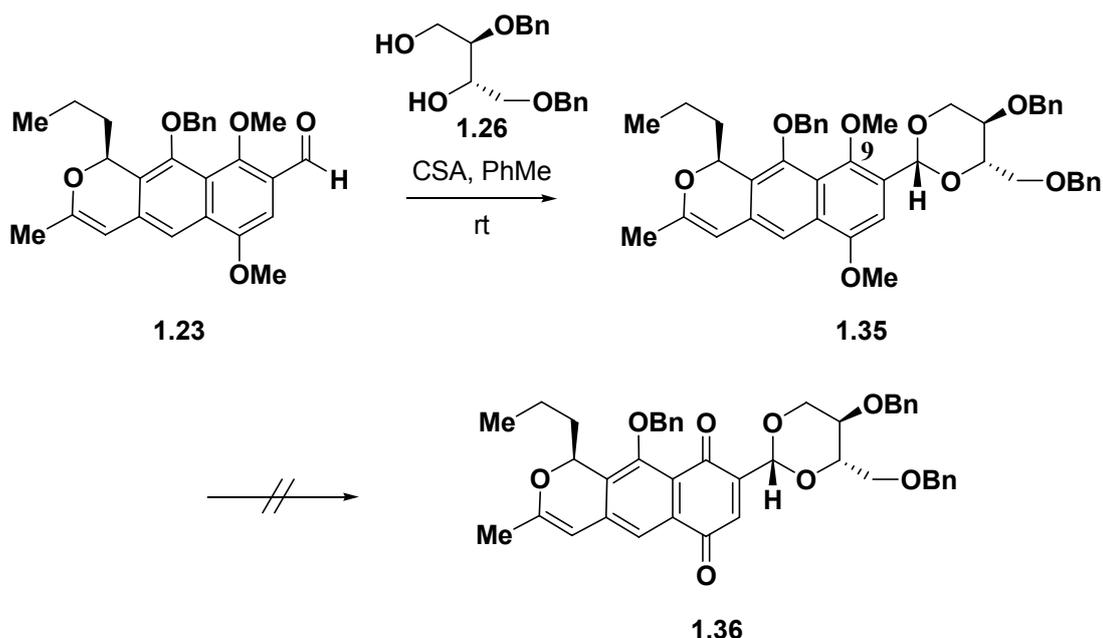
Scheme 1.10. Construction of 1,3-Dioxane Moiety and Investigation of the K1115 B₁ Skeleton.

As a result of these experiments, the dehydrated **1.35** possessing the 1,3-dioxane moiety had the necessity to convert a substrate easier to be oxidized. To fulfill the requirement, tetra-benzyl ether **1.43** would be determined to synthesize. Methyl group of **1.35** at C9 position was substituted to benzyl group and **1.35** would be changed into the substrate much easier to be oxidized by debenzylation. **1.43** was synthesized in a way similar with the methyl protected **1.35** as shown in Scheme 1.11.

There were three differences between Scheme 1.4 to 1.8 and 1.11: 1) tandem Michael-Dieckmann and subsequent aromatization proceeded efficiently in one-pot, increasing its yield. It was essential to regulate its reaction temperature. 2) Two kinds of stereochemistry, (1*R*)- and (1*S*)- configurations, at C1 position could not be enantiodivergently introduced from the compound **1.6**, and only (1*S*)-diastereomer could be synthesized. The reactivity of the benzyl protected **1.6** for Grignard reaction was largely changed to give the debenzylated product **1.45** in the case of *n*-PrMgBr, the ring-opening product **1.46** in the case of allylMgBr, and so on (Scheme 1.12). These undesired products could not be converted into the (1*R*)-diastereomer **1.47** or **1.48** by silane reduction. 3) The

stereoselective propylation was improved by changing the catalyst of the hydrogenation from Pd/C to Wilkinson complex. Wilkinson catalyst avoided producing over-reduced products, and made operations easier by purifying the lactol and the allylated product without column chromatography.
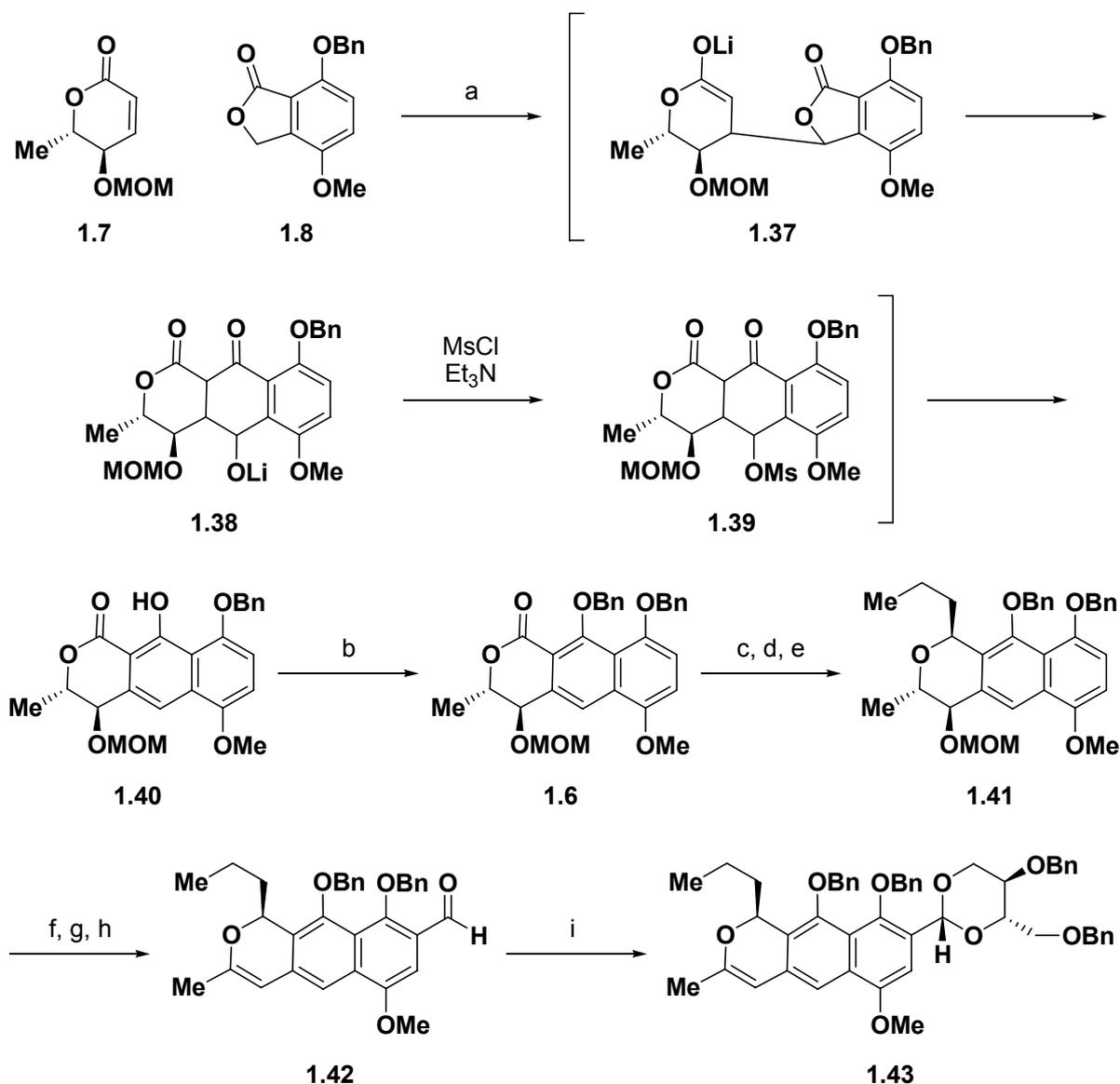
Scheme 1.11. Synthesis of the Compound **1.43**. Reagents and conditions: (a) LHMDS, THF, -78 °C to rt, 1 h, then MsCl, Et₃N, -15 °C, 15 min.; (b) BnBr, K₂CO₃, 18-crown-6, acetone, reflux, 11 h, 89 % (2 steps); (c) DIBAL, PhMe, -78 °C, 1 h; (d) allyltrimethylsilane, TMSOTf, CH₂Cl₂, -78 °C, 5 min.; (e) H₂, Wilkinson complex, EtOH, rt, 1 h, 90 % (3 steps); (f) Cl₂CHOCH₃, SnCl₄, CH₂Cl₂, -90 °C, 5 min.; (g) HCl-MeOH, rt, 1.5 h, 52 % (2 steps); (h) Burgess salt, THF, 50 °C, 1.5 h, 71 %; (i) **1.26**, CSA, PhMe, rt, 1.5 h, 82 %
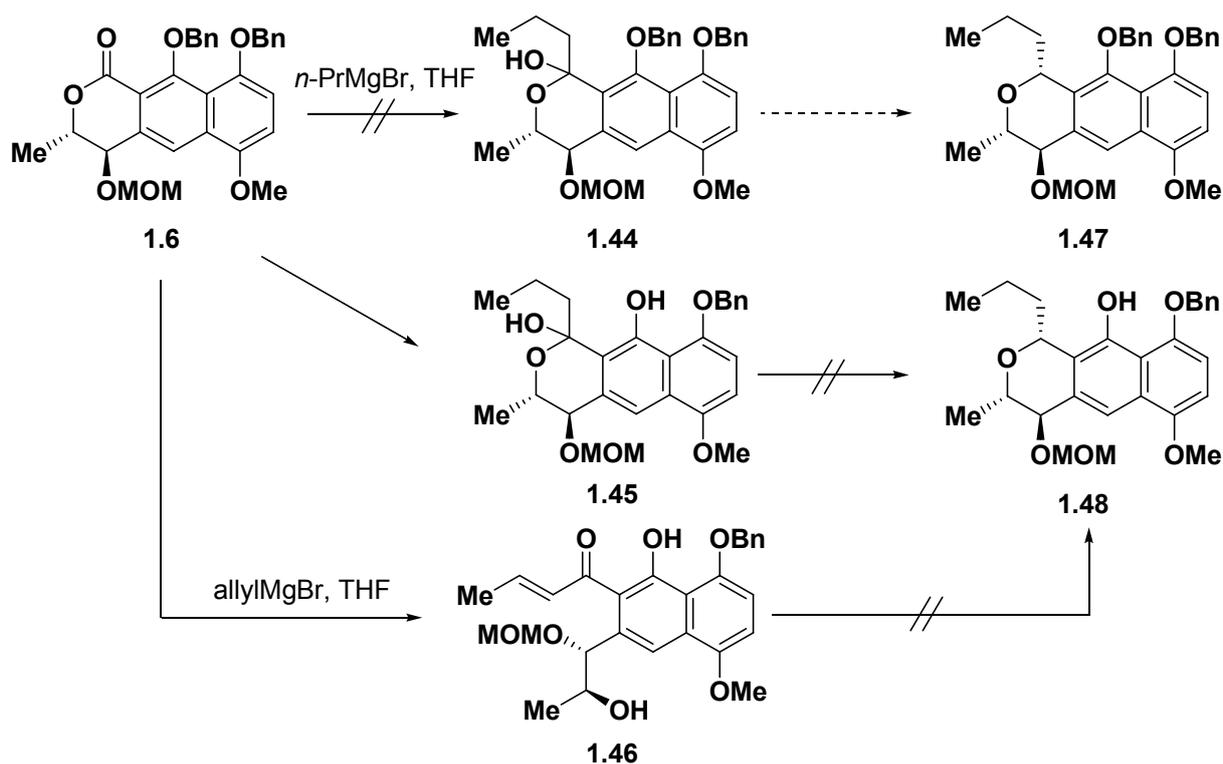
Scheme 1.12. Investigation of Synthesis of the (1*R*)-diastereomer **1.47** and **1.48**.

Reductive and oxidative cleavage of all benzyl groups of **1.43** was investigated (Scheme 1.13). However, all reductive conditions except for Raney Ni could not prevent the adsorption on catalysts and the cleavage of C1'-O bonds consisting of the 1,3-dioxane unit. In hydrogenation with the Raney Ni (W4), phenolic two of four benzyl groups were chemoselectively reduced to give **1.50**.

As a result of this experiment, if benzyl groups belonging to the 1,3-dioxane moiety were converted into other protective groups, it would be possible to construct the K1115 B$_1$ skeleton. The alternative protective group was determined to be TBS group, considering deprotecting conditions.
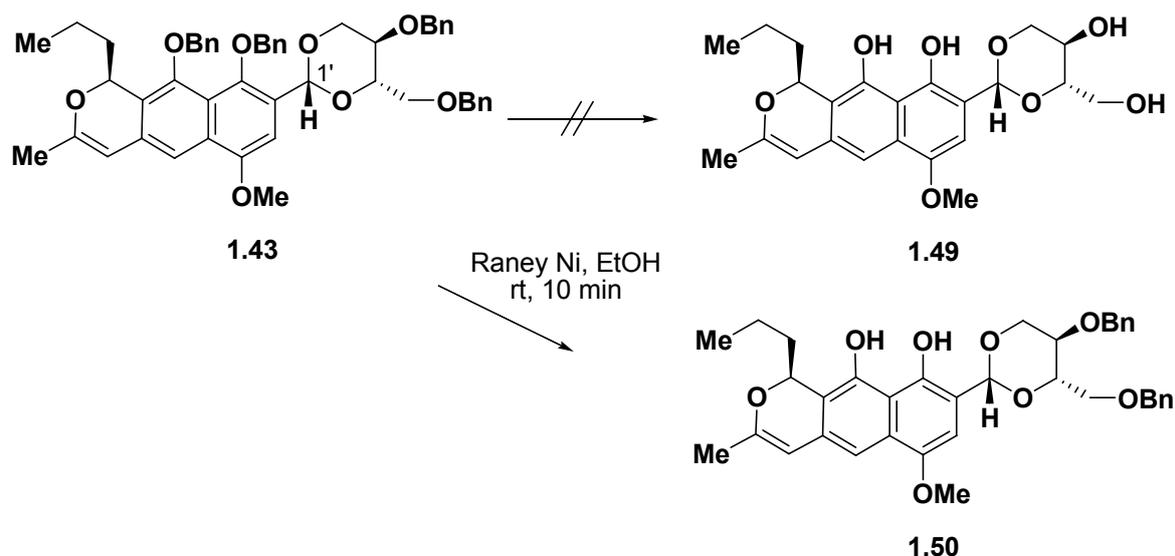
Scheme 1.13. Investigation of Deprotection.

Construction of the K1115 B_1 skeleton and total synthesis of (1*S*, 5'*S*)-(**1.2**) were achieved as shown in Scheme 1.14. Acetal formation of the dehydrated **1.42** using the TBS protected (5'*S*)-1,3-butane-diols **1.27** gave the acetal **1.51**. Hydrogenation with Raney Ni provided the corresponding dihydroxynaphthalene **1.52**, and after the celite filtration to remove the catalyst, treatment of the dihydroxynaphthalene **1.52** with salcomine under oxygen atmosphere afforded the *p*-quinone possessing the K1115 B_1 skeleton in only 13 % yield. After a number of experiments, the crude dihydroxynaphthalene **1.52** was found to be oxidized smoothly with PIFA into quinone mono acetal **1.54**, which was submitted to acid hydrolysis to give the naphthoquinone **1.53** in 62 % yield in 3 steps. Finally, deprotection of TBS group proceeded smoothly to give (1*S*, 5'*S*)-(**1.2**). And (1*S*, 5'*R*)-(**1.2**) was synthesized with (5'*R*)- 1,3-butane-diols ***ent*-1.27** instead of **1.27** by the .same method as that of (1*S*, 5'*S*)-(**1.2**).

Furthermore, (1*R*, 5'*R*)-(**1.2**) and (1*R*, 5'*S*)-(**1.2**) were synthesized from 3,4-di-*O*-acetyl-6-deoxy-D-glucal, the corresponding enantiomer of the starting material (Scheme 1.15).

Thus, all four diastereomers of K1115 B_1 were prepared.

18
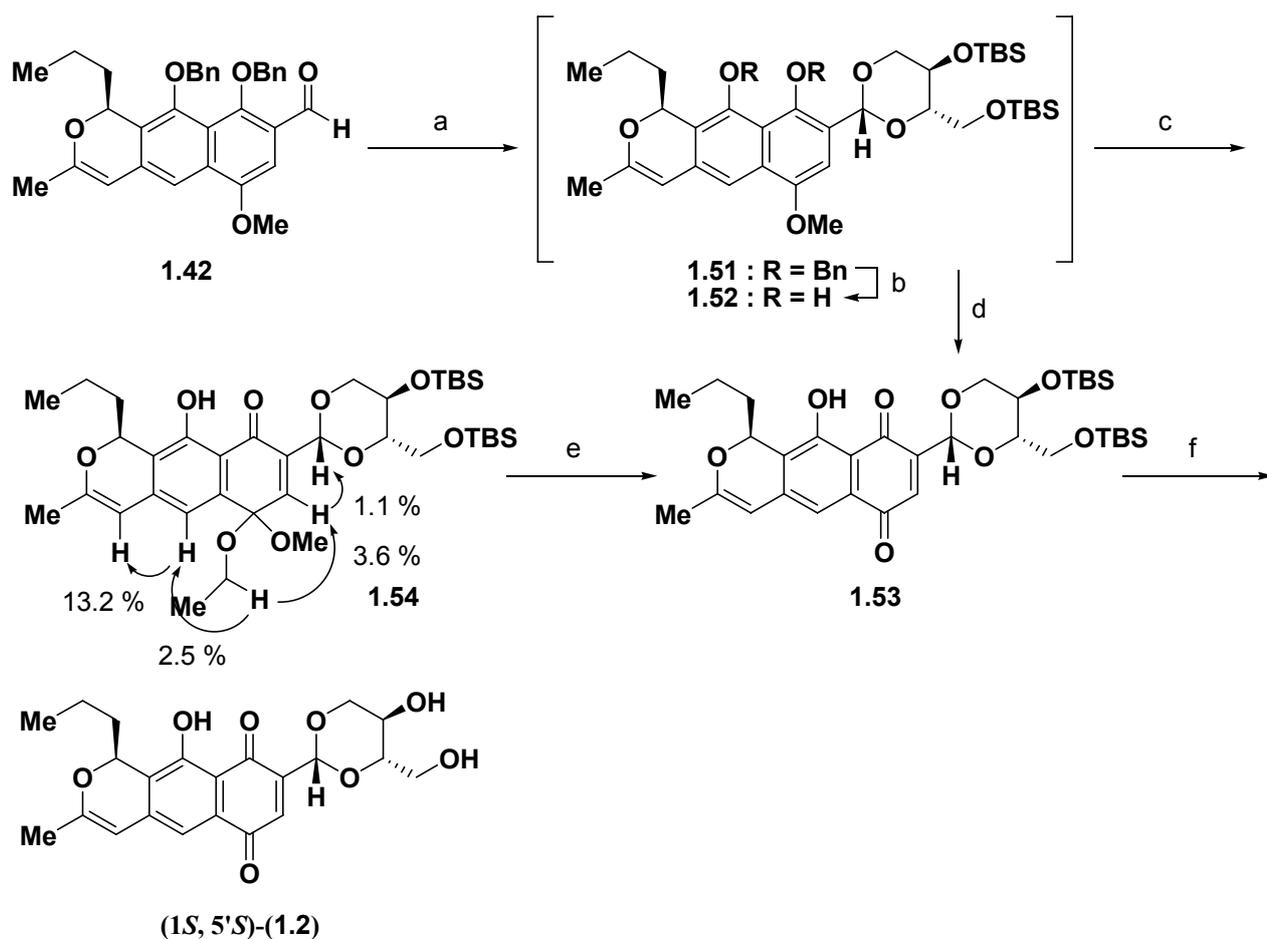
Scheme 1.14. Construction of the K1115 B$_1$ Skeleton and Total Syntheses of (1*S*, 5'*S*)-**1**. Reagents and conditions: (a) **1.27**, CSA, PhMe, rt, 1.5 h, 82 %; (b) Raney Ni, EtOH, rt, 10 min.;   (c) PIFA, 0 °C, 10 min; (d) O$_2$, salcomine, EtOH, rt, 1 h, 13 % (2 steps); (e) 50 % TFA, CHCl$_3$, 0 °C, 10 min, 62 % (3 steps); (f) TBAF, THF, 0 °C to rt, 20 min., 83 %
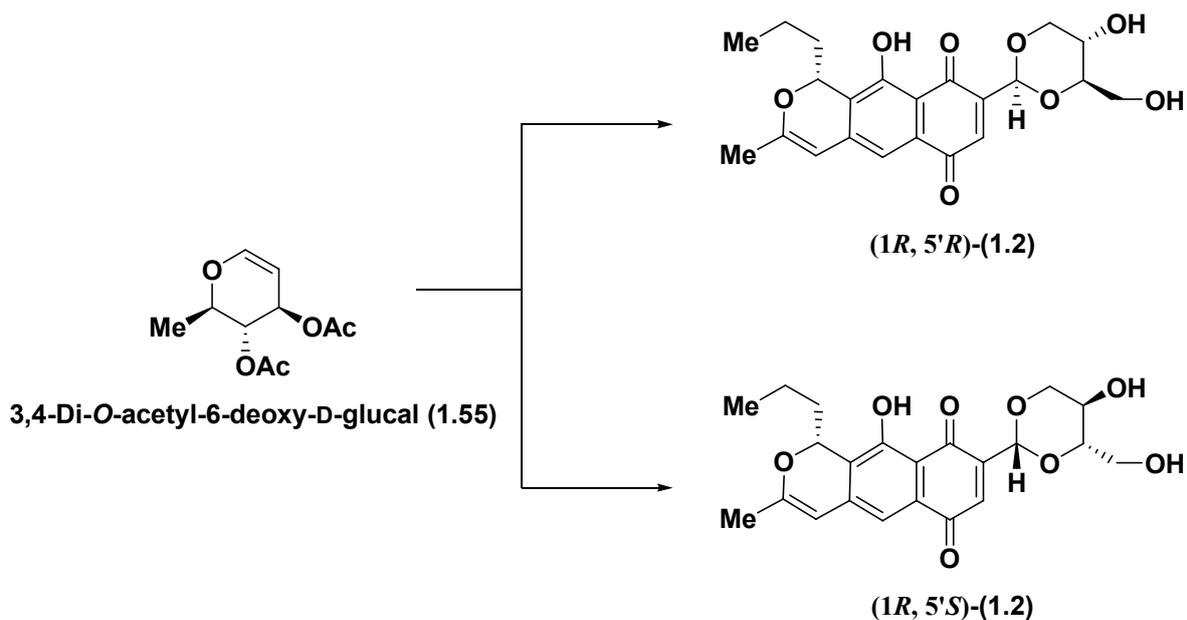
Scheme 1.15. Total Syntheses of (1*R*, 5'*R*)- (**1.2**) and (1*R*, 5'*S*)- (**1.2**).

Part 5. Structural Determination of Four Target Materials

The stereochemistry of the propyl group at C1 position and 1,3-dioxane moiety and the regioselectivity of the formyl group were determined by nOe experiments as shown in Figure 1.5, 1.6, and 1.7, respectively.

ent-**1.41**

Figure 1.5. Structural Determination of the Stereochemistry at C1 position by nOe Experiments.

1.56
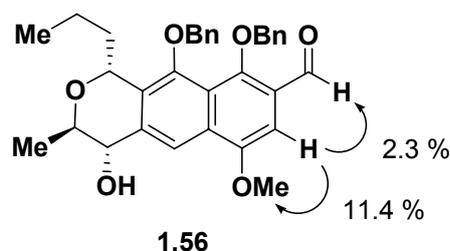
Figure 1.6. Structural Determination of the Regiochemistry of the Formyl Group by nOe Experiments.

(1*R*, 5'*S*)-(**1.2**)   (1*S*, 5'*S*)-(**1.2**)

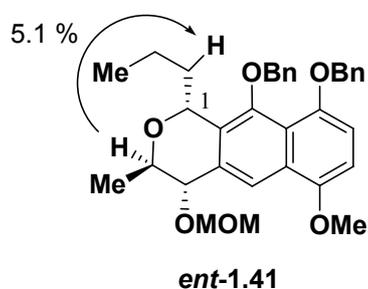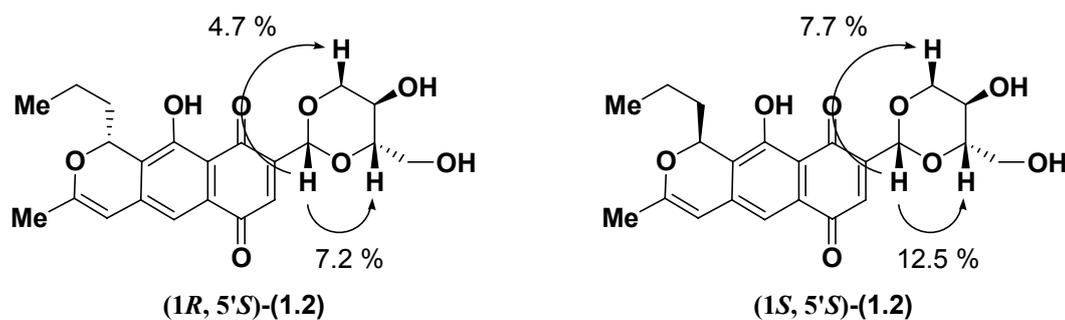**Figure 1.7.** Structural Determination of the Stereochemistry of 1,3-Dioxane Moiety by nOe Experiments.

Section 4. Total Synthesis and Structural Determination of K1115 B_1s (alnumycins)

It was not determined which diastereomer was K1115 B_1 by their spectral data and optical rotations since ^{1}H NMR spectra of four diastereomers were difficult to distinguish from that of natural product. The optical rotations of (1*S*, 5'*S*)- and (1*S*, 5'*R*)-(**1.2**) were levorotatory, while (1*R*, 5'*S*)- and (1*R*, 5'*R*)-(**1.2**) were dextrorotatory, respectively. The absolute values of the specific rotation of four diastereomers were around 1000.

Therefore, we isolated the natural product from the culture broth of *Streptomyces griseorubiginosus*, the strain of Eisai and Mercian group as follows. The strain of *Streptomyces griseorubiginosus* (Mer-K1115) was fermented by the modified Eisai's method: a will-sporulated slant of *Streptomyces griseorubiginosus* Mer-K1115 was used to inoculate 500 ml Erlenmeyer flasks each containing 50 ml of seed medium composed of glycerol 2.0 %, glucose 2.0 %, soybean meal 2.0 %, yeast extract 0.5 %, sodium chloride 0.25 %, calcium carbonate 0.32 %, and 0.2 % of metal salt solution containing 0.25 % copper sulfate, 0.25 % manganese chloride, and 0.25 % zinc sulfate, the pH being adjusted to pH 7.4 before sterilization. The seed flasks were incubated for 3 days at 28 °C on a rotary shaker at 220 rpm and 5 ml portions of the culture were transferred into 500 ml Erlenmeyer flasks containing 50 ml of production medium with the same composition as the seed medium, except that potato starch 2.0 % was used instead of glycerol 2.0 %. Fermentation was carried out for 2 days at 28 °C on a rotary shaker at 220 rpm. The harvested broth 2.0 l. was stirred with ethyl acetate 2.0 l. for 70 minutes. The liquid phase was separated from the mycelia mass by centrifungation (8000rpm, 4 °C, 20 minutes), and extracted with EtOAc. The organic layer was dried over anhydrous sodium sulfate, filtrated, and evaporated. The residue 1.43 g was purified by subsequent column chromatography containing silica gel column chromatography (hexane : 2-propanol = 4 : 1 including 1 % AcOH), LH-20 chromatography (MeOH), and silica gel column chromatography (toluene : acetonitrile = 3 : 1) to give the natural product 27.9 mg.

All diastereomers and the natural product were in hand, and the spectral data were compared with those of natural product. By comparison of ^{1}H NMR, it was found that the natural product contained two compounds as diastereomeric mixture. The mixture composed of 1.25:1 ratio of the compounds, which have been named K1115 $B_{1\alpha}$ and K1115 $B_{1\beta}$, respectively. K1115 $B_{1\alpha}$ was (1*S*, 5'*R*)- or (1*R*, 5'*S*)-(**1.2**), and K1115 $B_{1\beta}$ was (1*R*, 5'*R*)- or (1*S*, 5'*S*)-(**1.2**) (Figure 1.8).
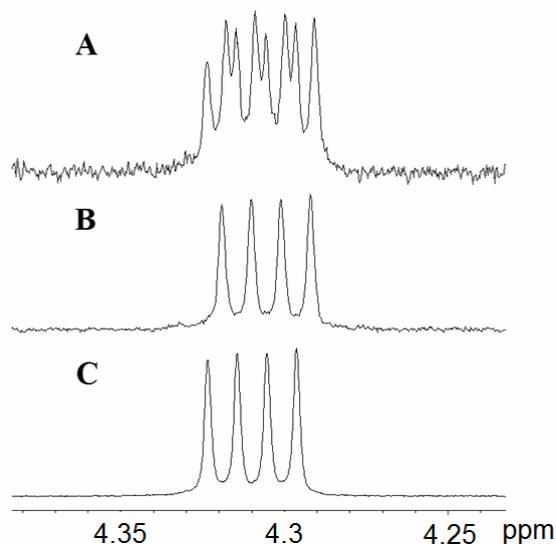
Figure 1.8. (A) ^{1}H NMR spectrum of natural K1115 B$_1$s. (B) ^{1}H NMR spectrum of (1R, 5'S)-(**1.2**). (C) ^{1}H NMR spectrum of (1S, 5'S)-(**1.2**).

The optical rotation of the natural K1115 B$_1$s was [α]$_D^{27}$ +1000° (c 0.1, MeOH). Therefore, the structures of K1115 B$_{1α}$ and K1115 B$_{1β}$ were determined to be (1R, 5'S)- and (1R, 5'R)-configuration, respectively.

And Bieber group reported that the optical rotaion of alnumycin was [α]$_D^{20}$ +170° (c 0.1, MeOH). It indicates that alnumycin may be another mixture of these four isomers.

Section 5. Conclusion

In conclusion, we have achieved the first total synthesis and the structural determination of K1115 B$_{1α}$ and K1115 B$_{1β}$ (alnumycins). Four diastereomers of K1115 B$_1$s, (1R, 5'R)-, (1R, 5'S)-, (1S, 5'R)-, and (1S, 5'S)-K1115 B$_1$s were synthesized. By comparison of 1H NMR spectra and the optical rotations, the absolute structure of K1115 B$_{1α}$ and K1115 B$_{1β}$ were determined to be (1R, 5'S)- and (1R, 5'R)-configuration, respectively. And alnumycin may be another mixture of these four isomers.

Chapter 2. The First Total Synthesis and the Structural Determination of TMC-66.

Section 1. Introduction

TMC-66 (**2.1**) was isolated from the fermentation broth of *Streptomyces* sp. A5008 as a new endothelin converting enzyme (ECE) inhibitor (Figure 2.1)[14]. ECE catalyzes the conversion and constitutes a potential regulatory step for the production of endothelin[15]. Therefore, ECE inhibitors would be therapeutically useful for treatment of the diseases involving ECE such as hypertension. TMC-66 (**2.1**) has a highly selective inhibitory activity for ECE with an IC_{50} value of 2.9 µM. Structurally, TMC-66 (**2.1**) had a benzo[*a*]naphthacenequinone and an oxazolidine ring. And only its relative stereochemistry had been determined, yet. Although many natural products having a benzo[*a*]naphthacenequinone structure, such as WS79089A, B, C[16], benaphthamycin[17], and benanomicins/pradimicins[18] had been already reported, a polycyclic structure with an oxazolidine is rare, as the only precedent in nature is cervinomycin[19], a xanthon anti-mycoplasmal antibiotic. WS79089B and pradimicin had been reported to be ECE inhibitors, too. The benzo[*a*]naphthacenequinone structure might be responsible for exhibiting ECE inhibitory activity. Therefore, its structure and bioactivity attracted us into its total synthesis. Herein, we present the first total synthesis and structural determination of TMC-66.
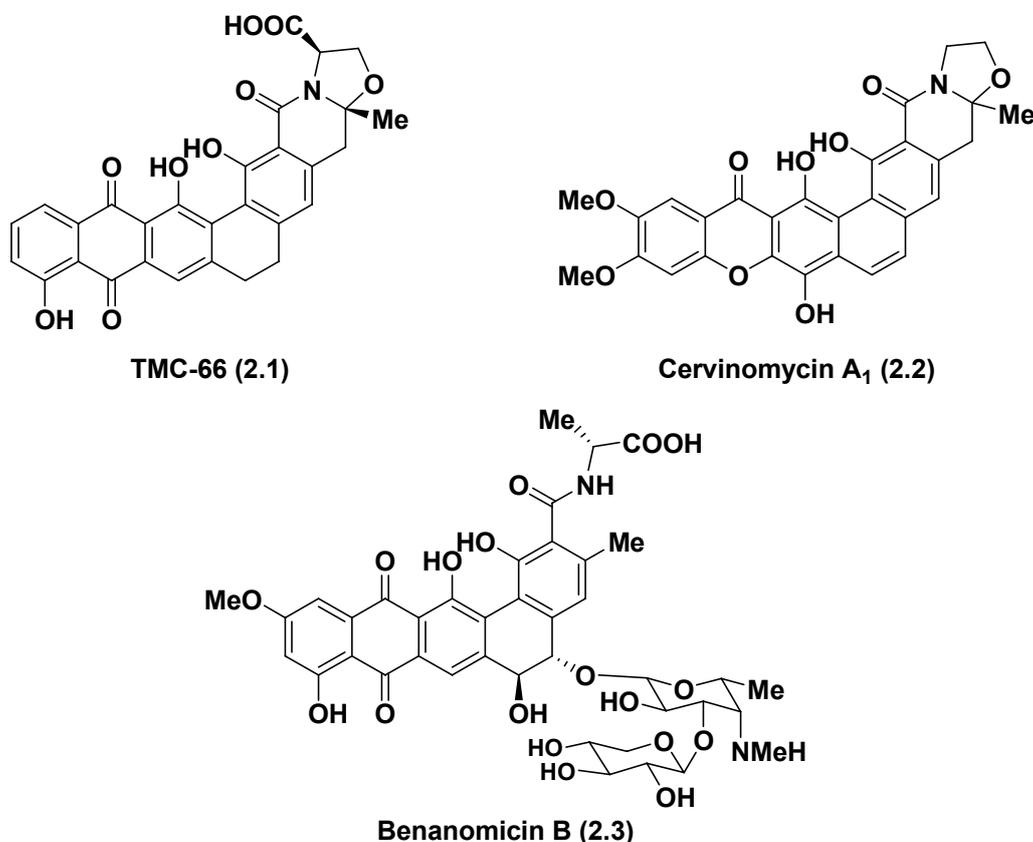
Figure 2.1. Natural products having a benzo[*a*]naphthacenequinone and structure of TMC-66.

Benzo[*a*]naphthacenequinones including benanomicins/pradimicins[20] and cervinomycins[21] have been synthesized. Their benzo[*a*]naphthacenequinone frameworks were constructed as follows.

Kelly's group accomplished the first total synthesis of (±)-cervinomycins A$_1$ (**2.2**) and A$_2$ (**2.11**) in 1989[21a] (Scheme 2.1). Xanthon fragment **2.6** was prepared by coupling between **2.4** and 2,6-diiodo-*p*-quinone (**2.5**). Phenol **2.7** was alkylated three times by three kinds of metalation to give the isocoumarin **2.8**. Condensation of **2.8** with ethanolamine produced isoquinolone fragment **2.9**. Pd-catalyzed arylation of styrene **2.9** with **2.6** furnished hexacyclic **2.10**, which was followed by the photocyclization to afford (±)-cervinomycins A$_1$ (**2.2**) and A$_2$ (**2.11**)
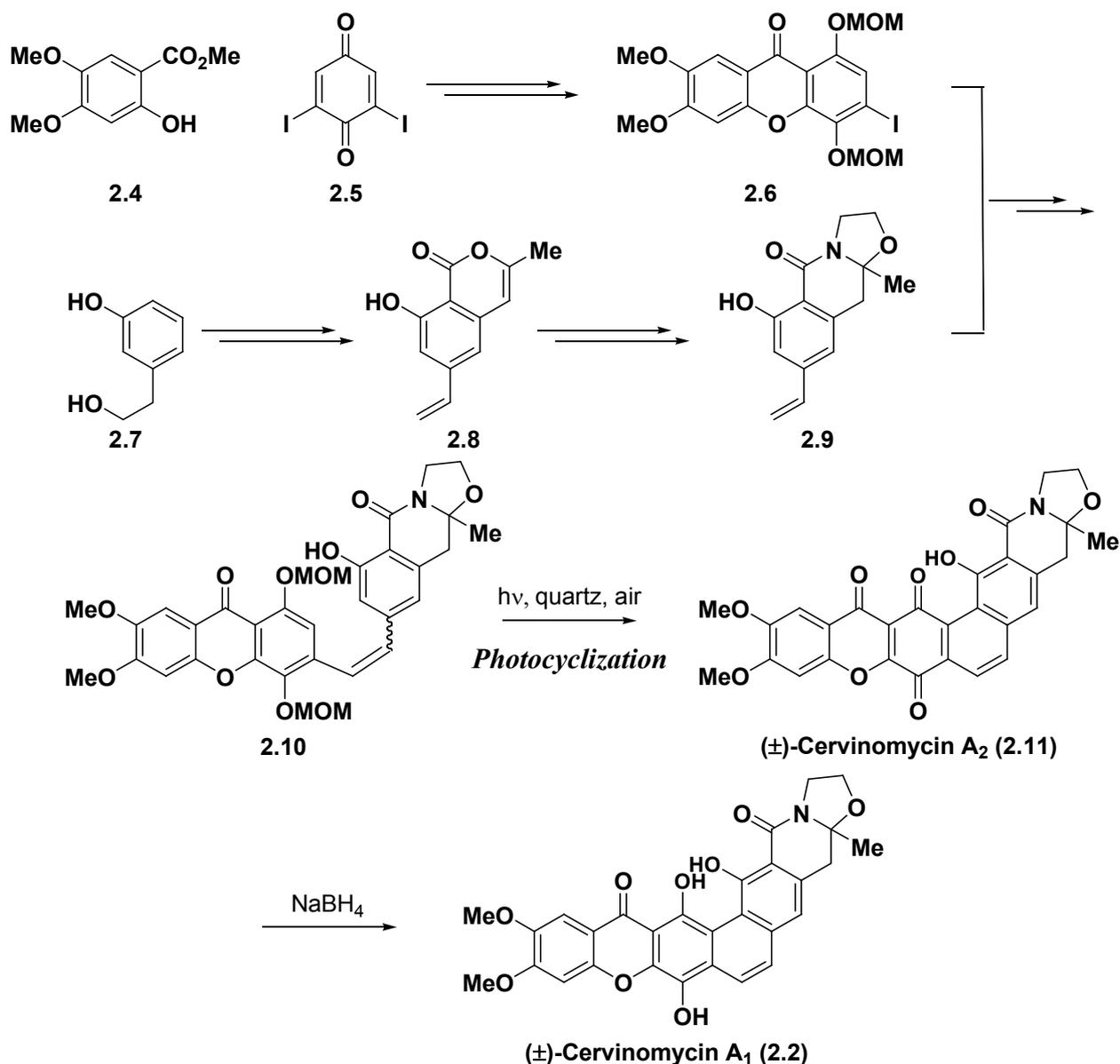
Scheme 2.1. The first total synthesis of (±)-cervinomycins A$_1$ and A$_2$.

Rao's group reported the total synthesis of (±)-cervinomycins A$_1$ (**2.2**) and A$_2$ (**2.11**) in 1991[21b] (Scheme 2.2). Naphthalene **2.12** was chosen as the starting material having the corresponding central ring in cervinomycin framework. After conversion of **2.12** to **2.13** through Friedel-Crafts reaction, ring extension by alkylation gave the tetracyclic compound **2.14**. Connection of **2.14** with **2.4** constructed the xanthon skeleton of ketoester **2.15**. Finally, condensation of **2.15** with ethanolamine furnished (±)-cervinomycins A$_1$ (**2.2**) and A$_2$ (**2.11**).
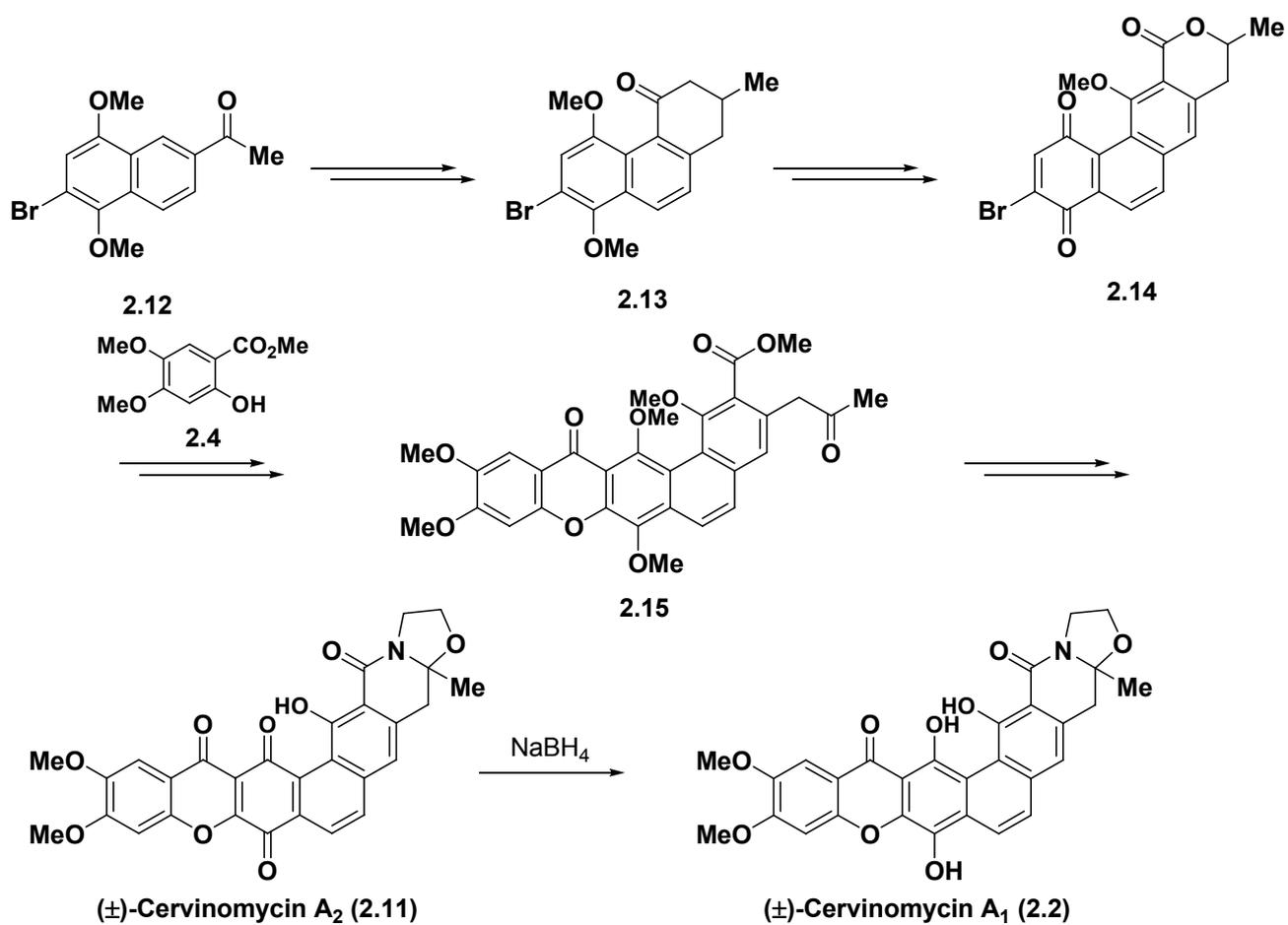
Scheme 2.2. Total synthesis of (±)-cervinomycins A$_1$ and A$_2$.

Krohn's group reported the biomimetic total synthesis of the benzo[*a*]naphthacenequinone, 5,6-dideoxypradione (**2.19**)[20b)] (Scheme 2.3). Diels-Alder reaction between **2.16** and **2.17** gave tricyclic compound, which was submitted to the extension of the side chains at the vicinal position by alkylation and Stille coupling to provide the triketo ester **2.18**. The mild base-catalyzed aldol cyclizations proceeded smoothly to afford 5,6-dideoxypradione (**2.19**).
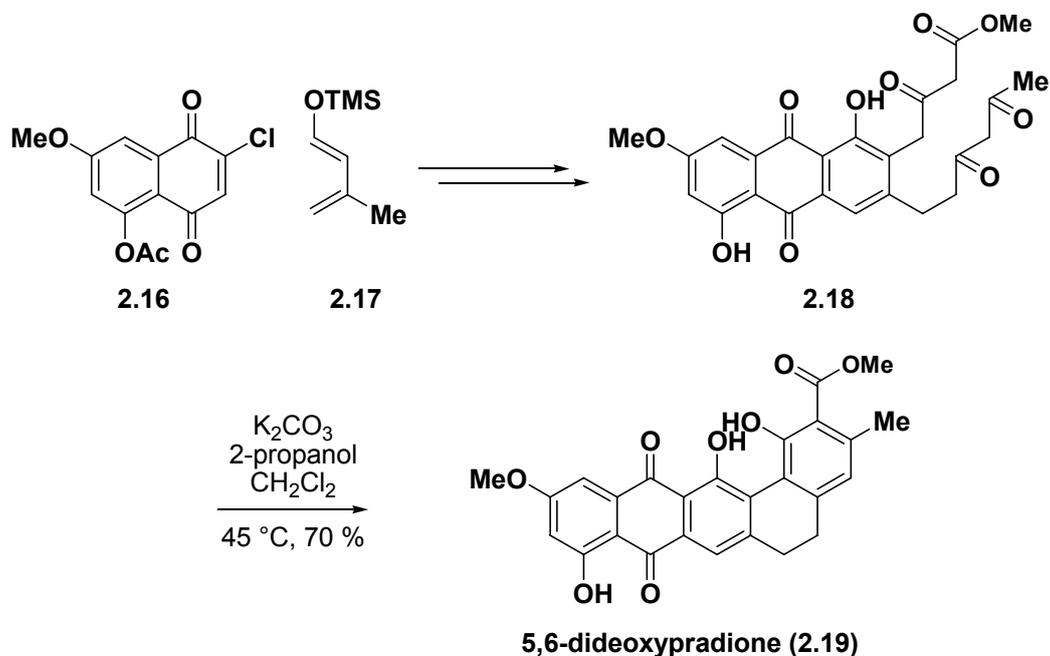
Scheme 2.3. Biomimetic total synthesis of 5,6-dideoxypradione (**2.19**).

Suzuki's group reported the first total synthesis of benanomicinone/pradimicinone in 1999[20e)], and benanomicin B (**2.3**) in 2005[20f)], based on a chiral transmission strategy using the semipinacol cyclization, which enabled the selective introduction of sugar moiety (Scheme 2.4). Yamaguchi esterification between carboxylic acid **2.20** and phenol **2.21** gave ester **2.22**, followed by Pd-mediated cyclization to afford lactone **2.23**. Lactone ring-opening by direct attack of D-valinol proceeded in 90 % yield with 91 : 9 diastereoselectivity to provide **2.24**. After the conversion of **2.24** to acetal-aldehyde **2.25**, the semipinacol cyclization in the presence of 1 equiv. of MeOH afforded the *trans*-adduct **2.26** with complete chiral transmission in 95 % yield. Finally, introductions of D-alanine and sugar moiety led to benanomicin B (**2.3**).
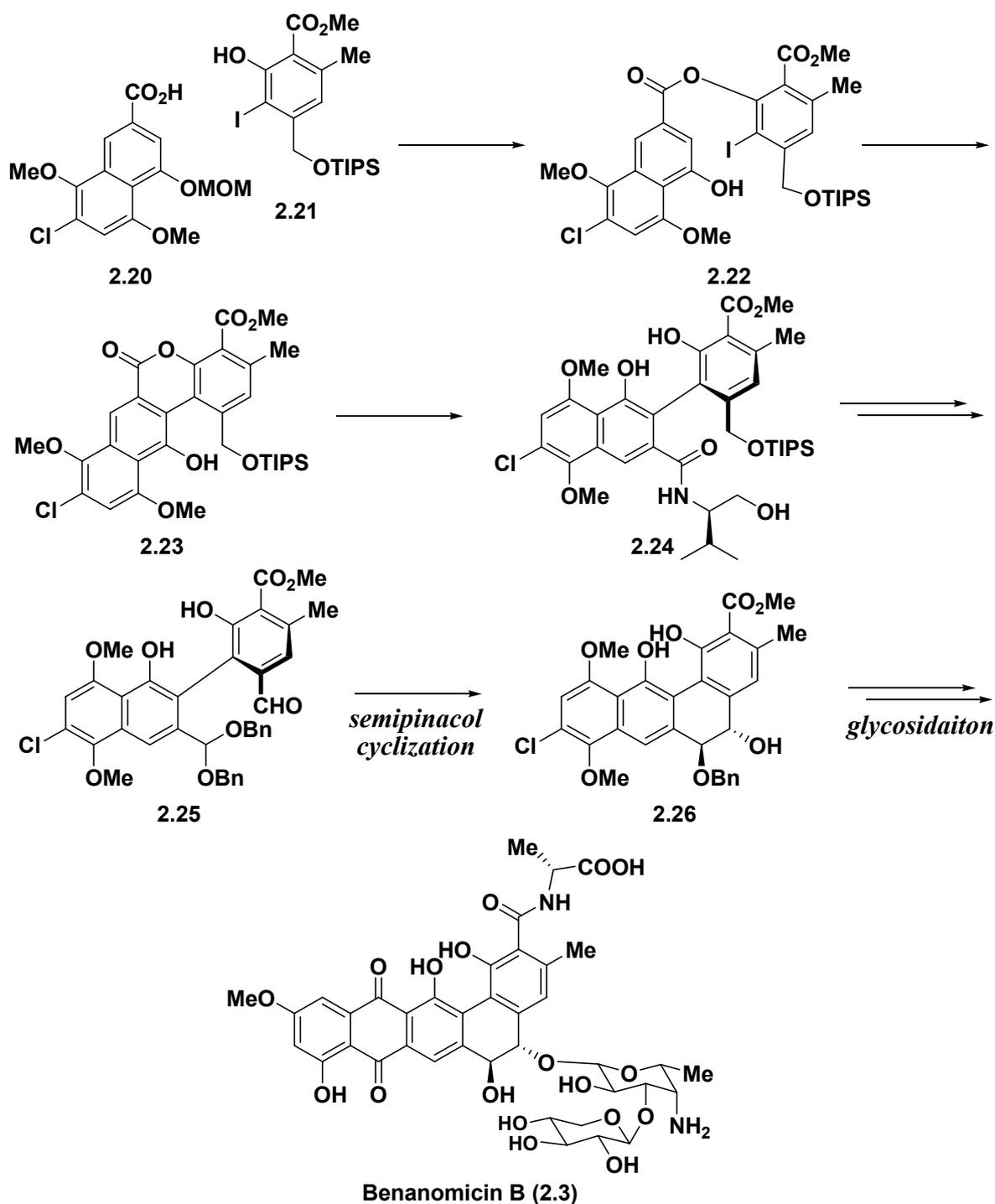
Scheme 2.4. The first total synthesis of benanomicin B.

On the basis of those previous papers, we determined that our total synthesis had to include an enantioselective condensation, a highly convergent route, and a highly efficient connection of segments.

Section 2. Retrosynthetic Analysis

Our synthetic plan is shown in Scheme 2.5. There were two points in our retrosynthetic analysis: 1) the intramolecular oxidative coupling of **2.27**, and 2) short-step synthesis of coupling segments **2.28** and **2.29** to establish efficiency and convergency.

The intramolecular oxidative coupling between C14a and C14b of **2.27** should be the key step to construct the benzo[*a*]naphthacenequinone skeleton of TMC-66 (**2.1**). This oxidative coupling would be challenge because of the different electron-withdrawing groups attaching to each phenol. Its regioselectivity was planned to be controlled by its own steric repulsion in the coupling mode. The hexacyclic **2.27** would be synthesized using Sonogashira coupling between similar-size segments **2.28** and **2.29**.

The oxazoloisoquinoline segment **2.28** would be afforded by stereoselective condensation of ketoester **2.30** with D-serine derivative. The anthraquinone segment **2.29** was designed to be synthesized from compound **2.31** by Diels-Alder cycloaddition.
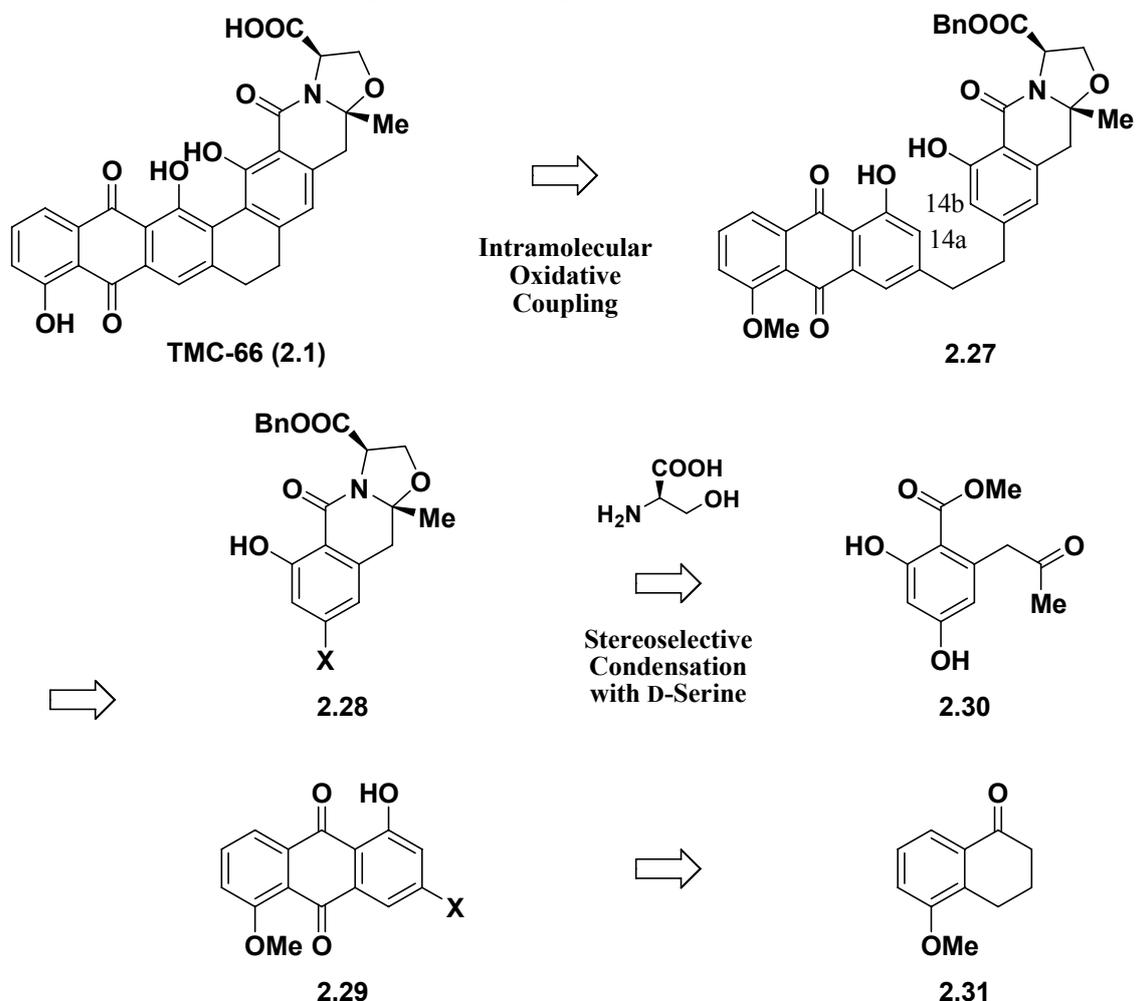
Scheme 2.5. Retrosynthetic analysis of TMC-66.

Section 3. Results and Discussion

Part 1. The Construction of the Anthraquinone Skeleton

The anthraquinone skeleton was constructed in two steps from 5-methoxy-1-tetralone (**2.31**) (Scheme 2.6). Oxidation of **2.31** under the oxygen atomosphere[22] and subsequent acetylation gave naphtoquinone **2.32**. Diels-Alder cycloaddition between **2.32** and **2.33** was followed by aromatization to construct the anthraquinone skeleton. The anthraquinone **2.3** was obtained by regioselective sulfonylation of **2.34**. Sonogashira coupling and subsequent desilylation provided the other anthraquinone segment **2.37**.
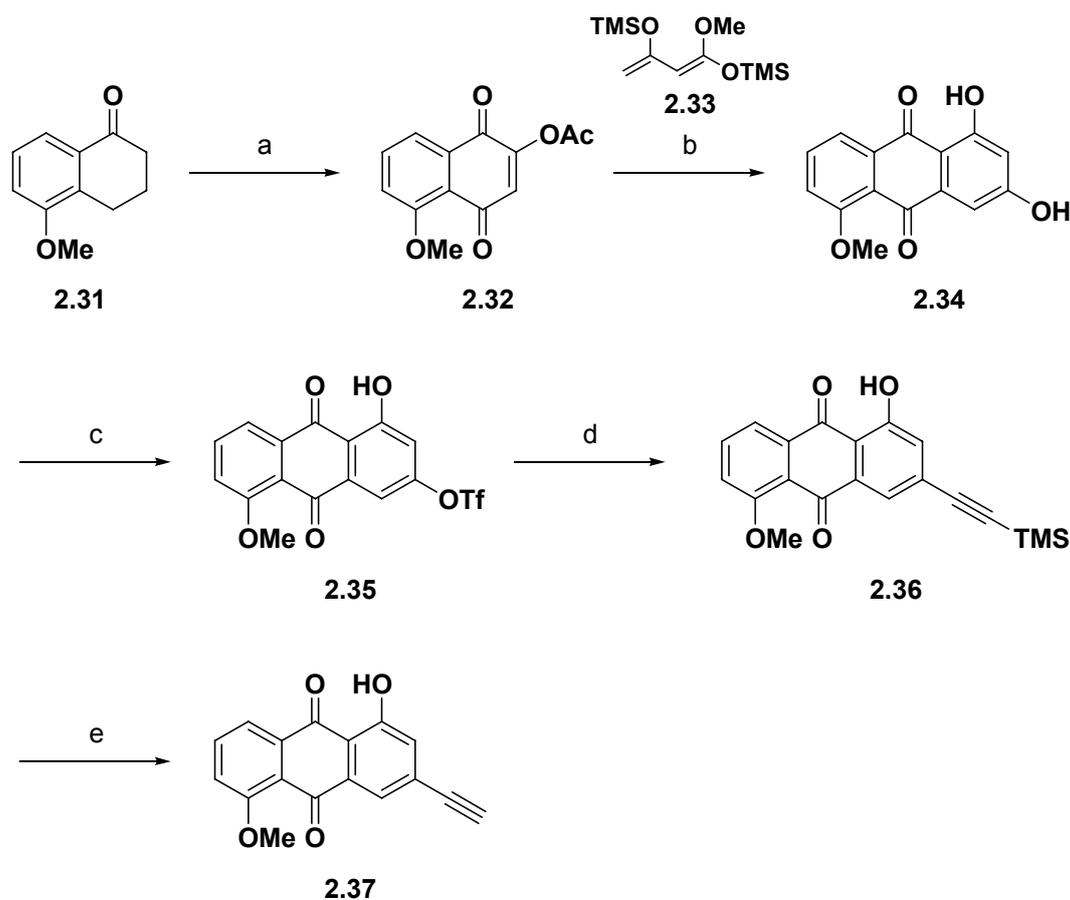
Scheme 2.6. Synthesis of anthraquinone segments. Reagents and conditions: (a) O_2, t-BuOK/t-BuOH, rt, 45 min, then Ac_2O, rt, 2 h, 47%; (b) **2.33**/toluene, 110 °C, 23 h, then pyridine, DMAP, 110 °C, 1.5 h, 70%; (c) TfCl, NHMDS, HMPA/THF, 0 °C, 5 min, 74%; (d) trimethylsilylacetylene, Pd(PPh$_3$)$_4$, CuI/ i-Pr$_2$NH-DMF, rt, 60 °C; (e) TBAF/THF, 0 °C.

Part 2. The Construction of the Oxazoloisoquinoline Skeletone and Studies of the Mechanism of the Stereoselective Condensation with D-Serine

Efficient synthesis of ketoester **2.30** was necessary to prepare in order to supply efficient amount of oxazoloisoquinoline segment. We discussed the three kinds of methodologies for synthesizing ketoester **2.30**. The first one had Diels-Alder cycloaddition as a key step to construct its aromatic ring (Scheme 2.7). The second was started from 5-resorcinol (**2.42**), an aromatic compound, through methoxycarbonylation by halogen-metal exchange (Scheme 2.8). The third one was the most efficient route to establish the ketoester **2.30** by introducing the functionalities into the methyl benzoate derivative in turn using palladium-catalysts (Scheme 2.11).

In the first pathway, ketoester **2.30** was provided in 13.7% yield in 6 steps from ethyl acetoacetate (**2.38**). Protection and reduction proceeded smoothly to give the corresponding aldehyde. Knoevenagel condensation of malonic acid dimethyl ester with the aldehyde gave α,β-unsaturated ester **2.39**. Diels-Alder cycloaddition between diene **2.40** and α,β-unsaturated ester **2.39** and the subsequent acidic hydrolysis afforded cyclohexenone **2.41**. After decarboxylation, oxidation accompanying with demethylation using iodine caused aromatization to give the ketoester **2.30**.
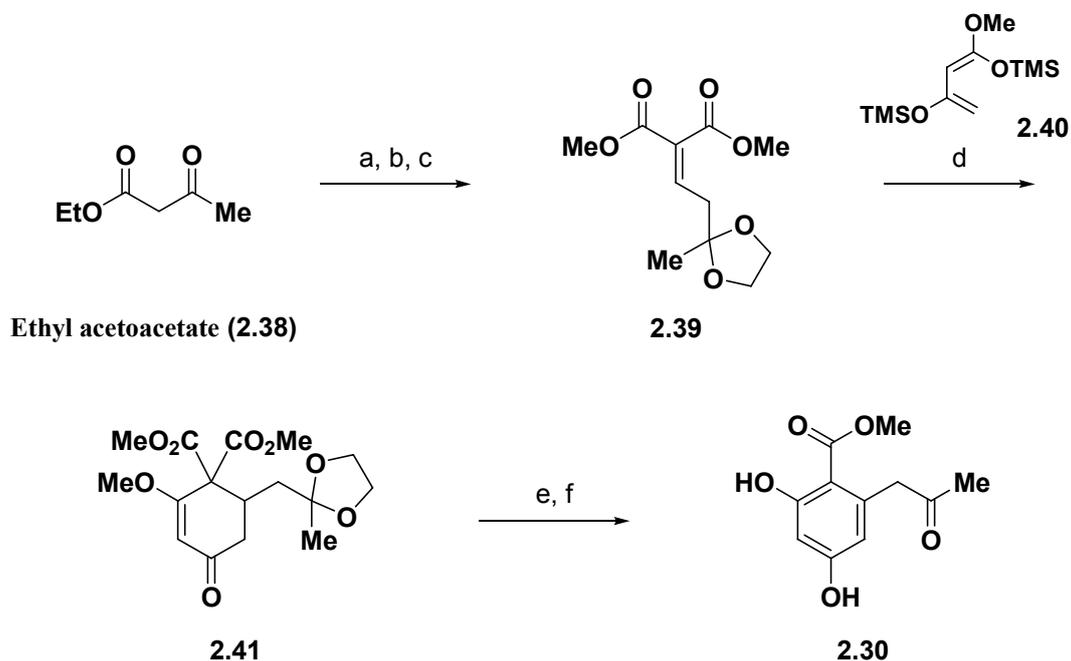
Scheme 2.7. The first generation synthesis of ketoester **2.30**. Reagents and conditions: (a) ethylene glycol, *p*-TsOH/benzene, reflux, 12 h, quant.; (b) DIBAL/CH$_2$Cl$_2$, -78 °C, 1 h, 87%; (c) malonic acid dimethyl ester, piperidine, AcOH/CH$_2$Cl$_2$, rt, 1 d, 70%; (d) **2.40**/toluene, then AcOH, 60 °C, 8 h, 54%; (e) NaCl, H$_2$O/DMSO, 130 °C, 12 h, 76%; (f) I$_2$, H$_2$O/THF, 40 °C, 7 h, 55%.

In the Scheme 2.7, there were two problems which we could not have solved. The first problem occurred in the Diels-Alder cycloaddition, the second did in aromatization. Each reaction proceeded in only moderate yield, and gave complex mixture involving hydrolysis byproducts, which could not converge on each objective compound any further. Thus, 5-resorcinol (**2.42**), an aromatic compound, was chosen as the new starting material.

In the second route, the ketoester **2.30** was obtained in 53.4% yield in 5 steps from 5-resorcinol (**2.42**). The second approach is prior to the first one, especially in the point of its yields. Methoxymethylation of two phenolic alcohol and regioselective bromination proceeded with almost quantitative. After halogen-metal exchange, addition of methyl chloroformate produced methyl ester **2.43** in 94% yield. Furthermore, acetylation at the benzyl position was accomplished with Weinreb amide **2.44** and the deprotonated methyl ester **2.43**. Finally, deprotection by acidic hydrolysis afforded the ketoester **2.30**.
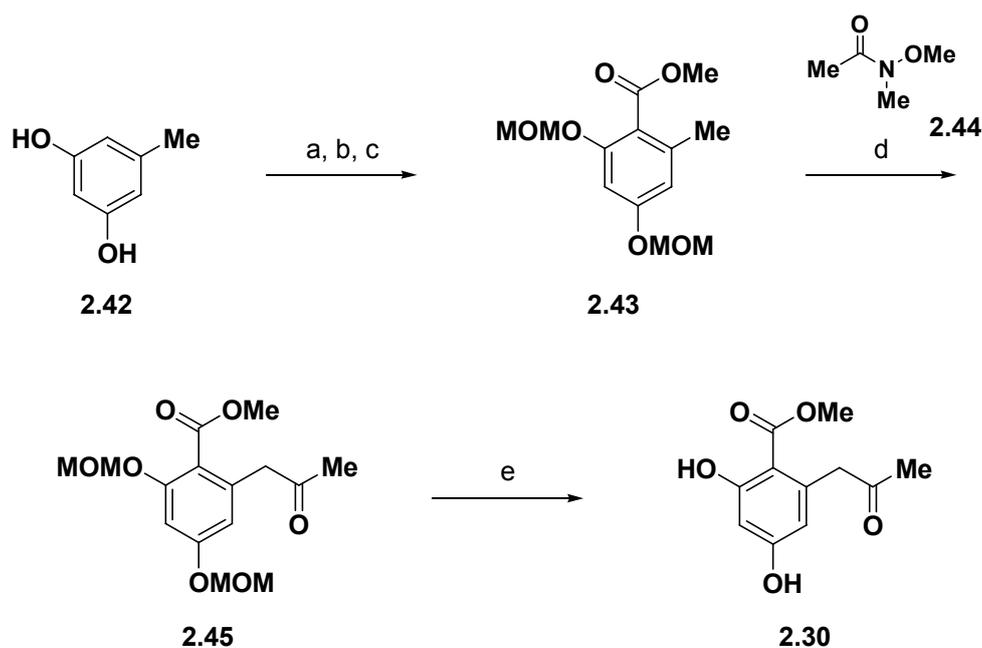
Scheme 2.8. The second generation synthesis of ketoester **2.30**. Reagents and conditions: (a) MOMCl, *i*-Pr$_2$NEt/DMF, rt, 1 h, 90%; (b) NBS/CHCl$_3$, 0 °C, 0.5 h, quant.; (c) ClCO$_2$Me, *sec*-BuLi/THF, -78 °C, 2 h, 94%; (d) LDA/THF, -78 °C, 4 h, 83%; (e) aq. HCl-MeOH, 50 °C, 10 h, 76%.

Next, the topic advanced to the stereoselective condensation and the synthesis of the oxazoloisoquinoline segments.

The condensation between the ketoester **2.30** and D-serine derivative **2.46** constructed oxazoloisoquinoline skeleton stereoselectively, and the subsequent triflation proceeded regioselectively to provide **2.47** (Scheme 2.9). Sonogashira coupling between **2.47** and trimethylsilylacetylene, followed by desilylation, gave the oxazoloisoquinoline segment **2.48**.
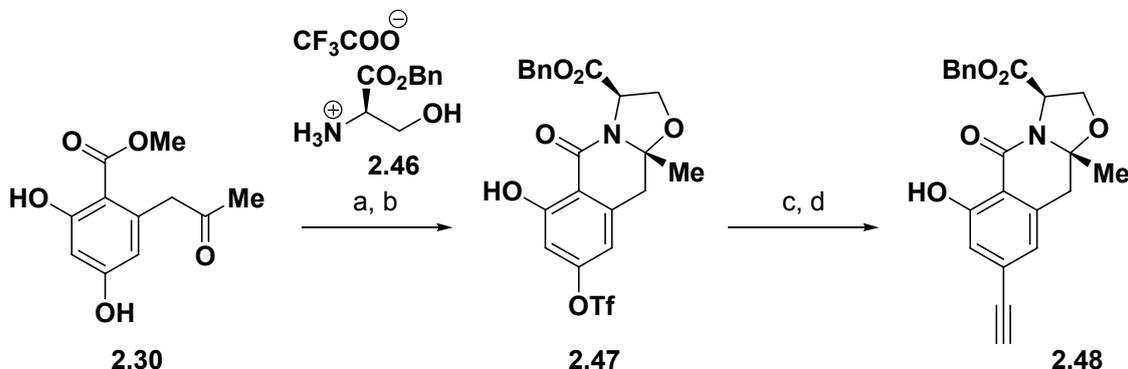
Scheme 2.9. Synthesis of oxazoloisoquinoline segments **2.47** and **2.48**. Reagents and conditions: (a) **2.46**, Et₃N, BnOH/THF, rt, 2 d, 79%; (b) TfCl, *i*-Pr₂NEt/CH₂Cl₂, 0 °C, 5 min, 72%; (c) trimethylsilylacetylene, Pd(PPh₃)₄, CuI/*i*-Pr₂NH-DMF, rt, 7 h, 67%; (d) TBAF/THF, rt, 7 h, 98%.

Although the oxazoloisoquinoline segment had been synthesized and the improvement of the route to ketoester **2.30** was achieved, the route was considered too long to achieve our goal, the efficient total synthesis. There were three kinds of issues in Scheme 2.8 and 2.9: 1) whether to start from a starting material possessing ester moiety in itself (four steps involved introducing methyl ester in Scheme 2.8: methoxymethylation, bromination, halogen-metal exchange, demethoxymethylation.). 2) whether to decrease steps for protection and deprotection. 3) self-condensation of **2.46** occurred.

On the basis of these ideas, the alternative retrosynthetic analysis was invented as shown in Scheme 2.10. Oxazoloisoquinoline segment **2.47** would be synthesized through the condensation of the new ketoester **2.49**. The ketoester **2.49** should be synthesized from a newly choiced starting material.
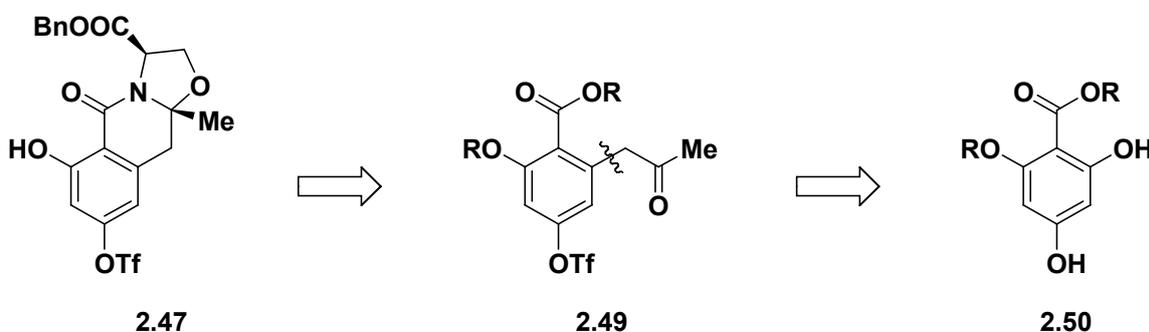
Scheme 2.10. The advanced retrosynthetic analysis of oxazoloisoquinoline segment **2.47**.

After a number of experiments, oxazoloisoquinoline segment **2.48** was obtained from acetonide **2.51** in 5 steps in 56.8% yield (Scheme 2.11). The protection of acetonide **2.51** produced bistriflate **2.52**. The regioselective Sonogashira coupling of **2.52** with trimethylsilylacetylene gave monotriflate **2.53** in quantitative yield. The triflate group of **2.53** was substituted to acetonyl group according to Migita's procedure in the presence of LiCl and Buchwald ligand to afford the ketoester **2.54**. Treatment of the ketoester **2.54** with a mixture of D-serine (1.2 equiv) and NaOMe (1.0 equiv.) in MeOH at 60 ℃ constructed oxazoloisoquinoline skeleton as a single diastereomer. Under these conditions, trimethylsilyl group was removed smoothly. The resulting carboxylic acid **2.55** was benzylated to give the oxazoloisoquinoline segment **2.48**.

In the stereoselective condensation, there was one device to prevent a side rection and epimerization of the cyclized product **2.55**. The device was to adjust the pH value to as neutral as possible. Benzoisopyranone **2.56** was found easy to be prepared as the major byproduct by the treatment of ketoester **2.55** with base. Under the previous conditions (Scheme 2.9), the similar side reaction occurred. Therefore, under the present conditions (Scheme 2.11), pH value was controlled to be about eight by mixing 1.2 equivalent of D-serine and 1.0 equivalent of NaOMe.
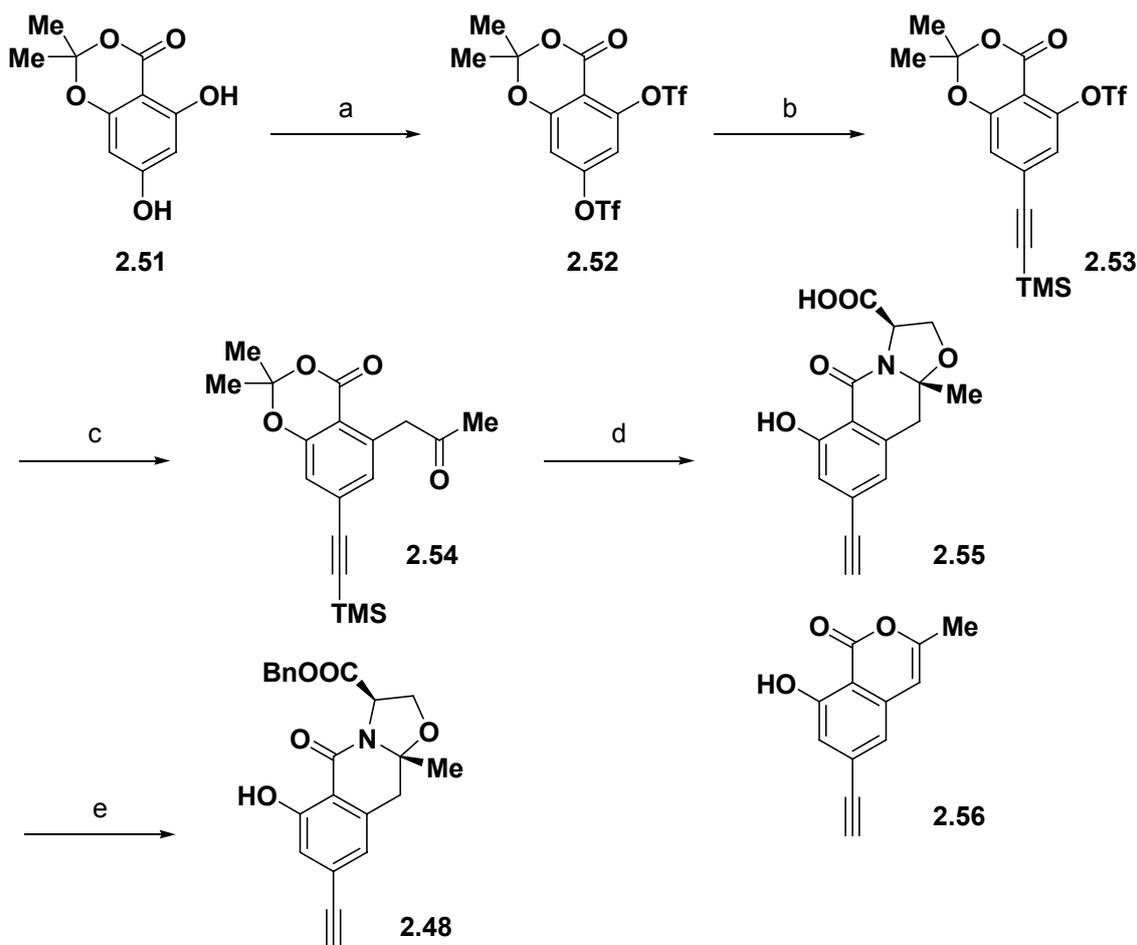
Scheme 2.11. The innovated synthesis of oxazoloisoquinoline segment **2.48**. Reagents and conditions: (a) Tf$_2$O/pyridine, 0 °C, 5 min, 95%; (b) trimethylsilylacetylene, PdCl$_2$(PPh$_3$)$_2$, CuI / *i*-Pr$_2$NH-toluene, rt, 5 min, quant.; (c) isopropenyl acetate, *n*-Bu$_3$SnOMe, Pd$_2$(dba)$_3$ · CHCl$_3$, LiCl, 2-diphenylphosphino-2'-(*N,N*-dimethylamino)biphenyl/toluene, 110 °C, 5 min, 83%; (d) D-serine, NaOMe/MeOH, 60 °C, 1 d; (e)BnBr, Cs$_2$CO$_3$/HMPA, rt, 12 h, 72% (2 steps).

The structural determination of oxazoloisoquinoline skeleton was determined by nOe of the tricyclic **2.48**. Methyl group of **2.48** was found to be *cis* to the benzyl ester group as shown in Figure 2.2. Thus, the stereochemistry of the C18 position was determined as (*R*)-configuration.
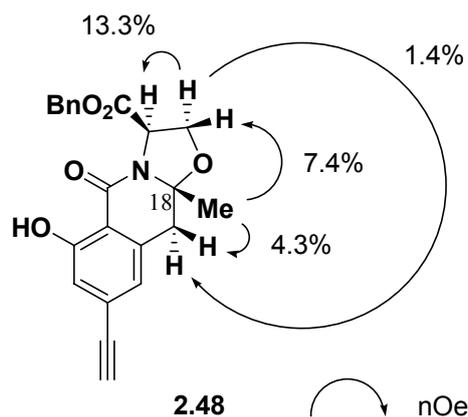
Figure 2.2. The structural determination of the oxazoloisoquinoline skeleton by nOe.

The mode of cyclization was determined by means of equations 1 and 2. Equation 1 showed 3 days were needed for benzopyranone **2.56** to react with D-serine to give the cyclized **2.55** under the same conditions as mentioned in Scheme 2.11. This result indicates that benzopyranone **2.56** was not the intermediate since the reaction time of the condensation with **2.54** was one day. Equation 2 meaned the amino group of D-serine under these conditions could not attack against the carbonyl group of lactone **2.57**. Accordingly, the mechanism of cyclization was explained as Scheme 2.12. The first step began from the attack of the amino group to the ketone group of ketoester **2.54**, followed by lactam formation and dehydration to give the cationic imine **2.58**. The final step was the intramolecular cyclization in a specific way to provide oxazoloisoquinoline **2.55**. The direction of the cyclization was controlled by the electrostatic and the steric repulsion between the carboxylate group and the lactam carbonyl group.

(eq. 1)

(eq. 2)
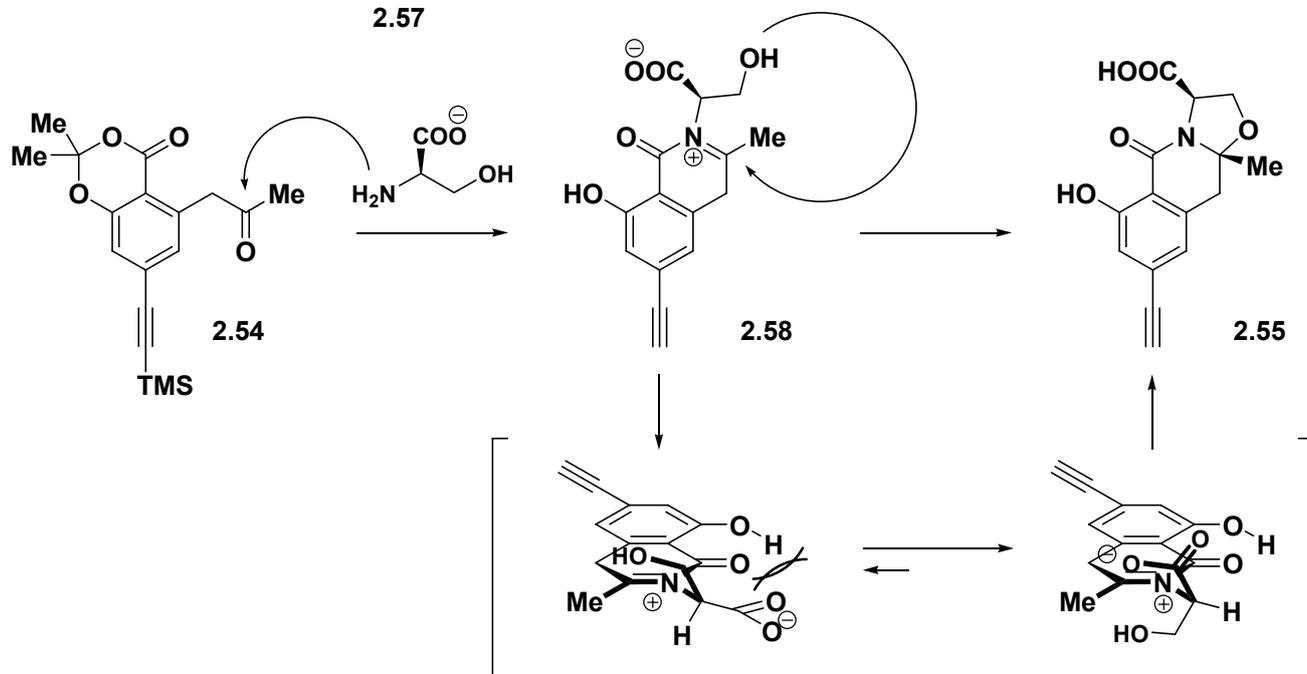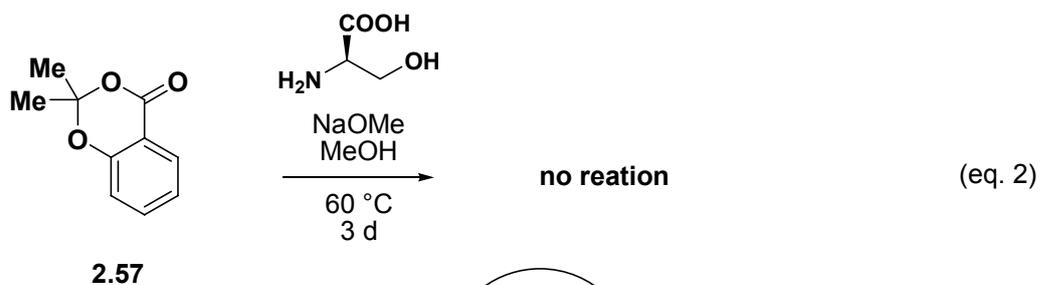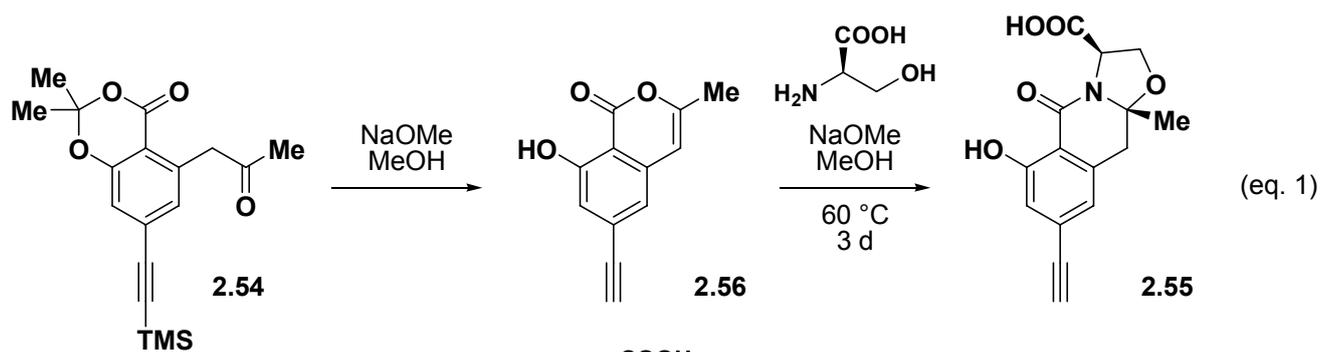
Scheme 2.12. The mechanism of the stereoselective condensation.

Part 3. The Intramolecular Oxidative Coupling and Total Synthesis of TMC-66

Both segments in hand, the connection between the segments **2.35** and **2.48** was accomplished by Sonogashira coupling to give hexacycle **2.59**, which was regioselectively reduced only at the acetylene moiety without cleavage of benzyl group to afford **2.60** (Scheme 2.13). The next intramolecular oxidative coupling was problematic. Among a variety of known oxidants, only the Koga's reagent CuCl(OH)‧TMEDA gave the desired product **2.61**, but in low yield (~30%). In order to change its reactivity, amine ligands were exchanged for other amines according to the Koga's procedure[X], which increased the yield of the coupling was improved to 89% when *N*-methylimidazole was used as the ligands. And the reaction proceeded completely regioselectively. Additionally, protection of the carboxylic acid of **2.60** was found to be essential to the intramolecular oxidative coupling. The free carboxylic acid derivative of **2.60** was decomposed under oxidative coupling conditions and did not produce the desired hepta-cyclic product. The structure of **2.61** was confirmed by nOe and HMBC as shown in Figure 2.3. Correlations of HMBC between H7 and C8, H12 and C13, and H4 and C19 were observed. Moreover, nOe was observed between H4 and H19. Thus, oxidative coupling product **2.61** should possess the TMC-66 skeleton. Finally, demethylation and debenzylation was realized by treatment of **2.61** with BBr$_3$ to afford TMC-66 (**2.1**). The spectral data of the synthetic TMC-66 (**2.1**) were identical with those of the natural product. The optical rotation of the reddish orange solution of the synthetic TMC-66 (**2.1**) was levorotatory, $[\alpha]_D^{26}$ -73° (*c* 0.09, CHCl$_3$), as that of the brown solution of the natural product ($[\alpha]_D^{24}$ -327° (*c* 0.01, CHCl$_3$)). Therefore, the total synthesis of TMC-66 was achieved to determine its absolute configuration as (16*R*, 18*R*).
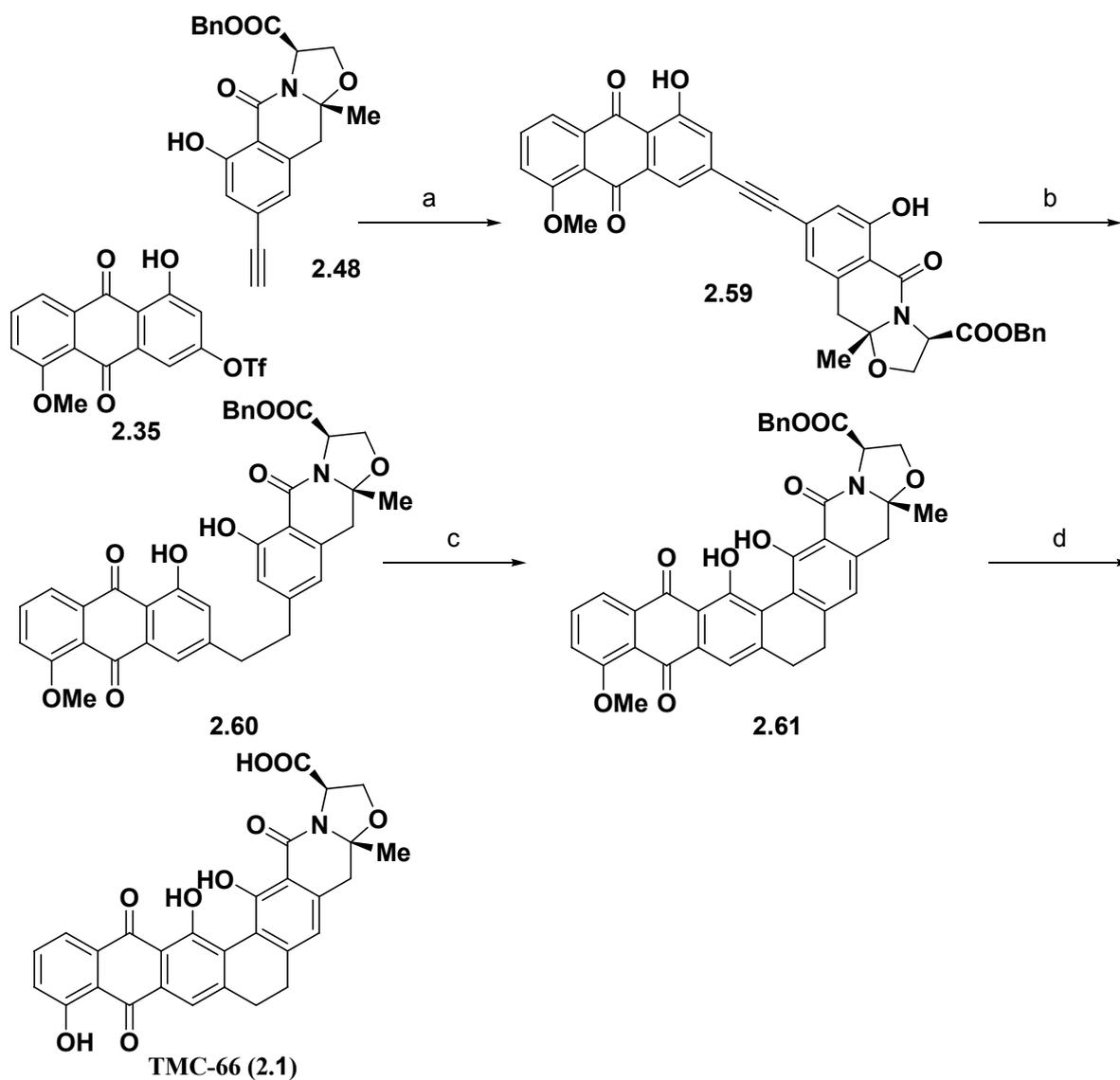
Scheme 2.13. Total synthesis of TMC-66 (**2**). Reagents and conditions: (a) Pd(OAc)$_2$, PPh$_3$, CuCl/i-Pr$_2$NH-DMF (1:5), rt, 5 min, 78%; (b) H$_2$/xylene, 120 °C, 50 min, 81%; (c) Cu-NMI complex/DMF, reflux, 1.75 h, 89%; (d) BBr$_3$, CH$_2$Cl$_2$, -78 °C, 30 min, 70%. NMI = N-methylimidazole.
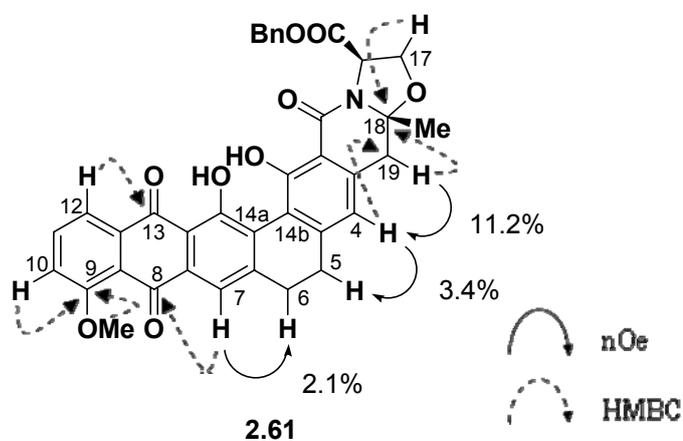
Figure 2.3. Sturactural elucidation of heptacyclic **2.61** by HMBC and nOe experiments.

The optical purity of the synthetic **2.1** was confirmed with a chiral 1,2-diphenylethane-1,2-diamine[X)] by ^{1}H-NMR (Figure 2.4). Racemic TMC-66 was synthesized from *dl*-serine as above. The spectrum A is ^{1}H-NMR spectrum of a mixture of racemic TMC-66 and (1*R*, 2*R*)-1,2-diphenylethane-1,2-diamine in the ratio of 2:1, while the spectrum B is that of a mixture of synthetic (-)-TMC-66 (**2.1**) and (1*R*, 2*R*)-1,2-diphenylethane-1,2-diamine. The spectrum A shows that (+)- and (-)-TMC-66 are distinguishable in the presence of a chiral 1,2-diphenylethane-1,2-diamine. The protons around the chiral centers, including H16 and H17, and C*H*-N of the chiral amine were observed between 4.15 and 4.75 ppm. Although they are broadened, these spectra are obviously discerned. The peaks around δ 7.75 ppm are corresponding to H12, H7, and H11. Even these protons are far from the chiral centers (C16 and C18) of TMC-66, they are also distinguishable between (+)- and (-)-TMC-66 with spectrum A. Obviously, the spectrum B show that the synthetic TMC-66 (**2.1**) was highly optical pure. Therefore, the difference of the value of the optical rotation was due to the inherent color of TMC-66 (**2.1**).
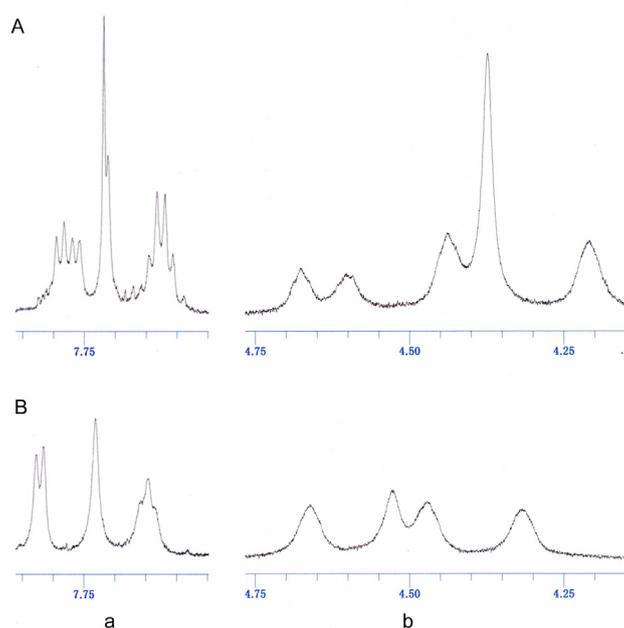
Figure 2.4. ^{1}H-NMR spectrum of (A): 2:1 mixture of racemic TMC-66 & (1*R*, 2*R*)-1,2-diphenylethane-1,2-diamine, and (B): 2:1 mixture of synthetic (-)-TMC-66 (**2.1**) & (1*R*, 2*R*)-1,2-diphenylethane-1,2-diamine. (From the left side) a: H12, H7, and H11. b: H16, C<u>H</u>-NH2 of diamine, H17a, and H17b.

Section 4. Conclusion.

In conclusion, the first total synthesis and structural determination of TMC-66 have been achieved. Additionally, the highly stereoselective condensation of ketoester **2.30** and D-serine and the regioselective intramolecular oxidative coupling having electron withdrawing groups were realized in high yield. The mode of cyclization have been disclosed. The efficient and stereoselective route to the benzo[*a*]naphthacenequinone fused with an oxazolidine ring possessing stereogenic centers has been established.

Chapter 3. Summary

Chapter 1. The First Total Synthesis and Structural Determination of K1115B$_1$s (alnumycins)

1. Tricyclic framework was efficiently constructed using tandem Michael-Dieckmann type reaction followed by aromatization in one-pot.

2. Propyl group at C1 position was stereoselectively introduced by using a sugar as its chiral resource.

3. Stereoselective construction of 1,3-dioxane moiety was accomplished by acetalization with chiral 1,3-butane-diol synthesized from D-glucose.

4. All four diastereomers of K1115B$_1$, (1R, 5'R)-, (1R, 5'S)-, (1S, 5'R)-, and (1S, 5'S)- K1115B$_1$s were synthesized convergently.

5. The natural product was isolated from the culture broth of *Streptomyces griseorubiginosus*, the strain of Eisai and Mercian group. The strain of *Streptomyces griseorubiginosus* (Mer-K1115) was fermented by the modified Eisai's method. It was found that the natural product contained two compounds as diastereomeric mixture. The mixture composed of 1.25:1 ratio of the compounds.

6. By comparison of ^{1}H NMR spectra and the value of the optical rotations, the absolute structures of K1115B$_{1\alpha}$ and K1115B$_{1\beta}$ were determined to be (1R, 5'S)- and (1R, 5'R)- configuration, respectively.

7. Alnumycin may be another mixture of four isomers.

Chapter 2. The First Total Synthesis and Structural Determination of TMC-66

1. The anthraquinone segment was synthesized in 3 steps using Diels-Alder cycloaddition as a key step.

2. The oxazoloisoquinoline segment was obtained in 5 steps in 56.8% yield by subsequent regioselective introductions of its functional groups followed by stereoselective condensation with D-serine.

3. Two segments were connected by Sonogashira coupling and the chemoselective hydrogenation of the acetylene moiety was accomplished to give the hexacyclic compound in high yield.

4. The regioselective intramolecular oxidative coupling was realized in high yield using Cu-NMI complex as an oxidant.

5. The first total synthesis and structural determination of TMC-66 have been achieved. The efficient and stereoselective route to the benzo[*a*]naphthacenequinone fused with an oxazolidine ring possessing stereogenic centers has been established. And its stereochemistry was determined to be (16*R*, 18*R*)-configuration.

Chapter 4. Experimental

Section 1. General Methods

^{1}H NMR spectra were recorded at 600 MHz with Bruker AVANCE 600 and 400 MHz with JEOL ECS 400 instrument. Coupling constants (J) are reported in Hz. ^{13}C NMR spectra were recorded at 150 MHz with Bruker AVANCE 600 instrument. Chemical shifts (δ) are quoted in parts per million (ppm) and referenced to the residual solvent peak. Melting point (mp) determinations were performed by using a Yanako MP-S3 instarument. FT-IR spectra were recorded at JEOL JIR-WINSPEC 50. HRMS and MS were obtained with a JEOL JMS-SX102A and JEOL JMS-GCMateII. Optical rotations were measured with a JASCO DIP-370 and P-2200. All reactions were monitored by analytical thin layer chromatography which was performed on 0.25 mm E. Merck silica gel plates (60F$_{254}$) using UV light as visualizing agent, an ethanol solution of p-anisaldehyde, an ethanol solution of sodium phosphorous molybdate, and heat as a developing agent. Flash and column chromatography was performed using Kanto Kagaku Ltd. Silica gel (60N neutral, particle size 0.040-0.063 mm) and Fuji Silysia Chemical Ltd. Silica gel (BW-820MH, particle size 6nm), respectively.

Section 2. Materials

All reactions were carried out under an argon atmosphere with dry, freshly distilled solvents under anhydrous conditions, unless otherwise noted. THF was distilled from sodium/benzophenone ketyl. Dichloromethane was distilled from phosphorous oxide. Acetonitrile, benzene, 1,2-dichloroethane, DMSO, methanol, and toluene were distilled from CaH$_2$. DMF was distilled from CaH$_2$ after drying by CuSO$_4$. And all other reagents were purchased at the highest quality from Aldrich, Kanto Chemical Co. Ltd., TCI, Wako, and so on.

Section3. The First Total Synthesis and the Structural Determination of antibiotics K1115 B₁s (alnumycins).

7-(Benzyloxy)-4-methoxyisobenzofuran-1(3*H*)-one (1.8)
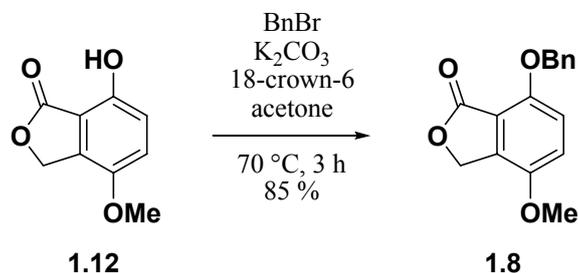
To a solution of the compound **1.12** 7.93 g (45.0 mmol) in acetone 320 ml were added 18-crown-6-ether 13.1 g (49.6 mmol), potassium carbonate 12.3 g (89.0 mmol), and benzyl bromide 8.10 ml (68.1 mmol). After reflux at 70 °C for 3 hours, the reaction mixture was evaporated in vacuo. To the resulting residue was added H₂O 300ml, followed by extraction with ethyl acetate (3×300 ml). After evaporation in vacuo, the residue was purified by flash column chromatography (toluene-ethyl acetate 8:1) and recrystallization from a mixture of hexane and chloroform to yield the compound **1.8** as a colorless needle (10.2 g, 38.3 mmol, 85 %).

R_f: 0.45 (toluene-EtOAc 6:1)

HR-MS (FAB+): calcd. for $C_{16}H_{15}O_4$: 271.0970, found 271.0987 [M+H]⁺.

mp: 142.5-143.0 °C

IR (cm⁻¹, KBr): 3029, 1760, 1507, 1277, 1014.

¹H NMR (CDCl₃, 600MHz) δ (ppm): 3.82 (3H, s, -OMe), 5.18 (2H, s, H-9), 5.26 (2H, s, -O-C*H₂*-Ph), 6.85 (1H, d, *J* = 9.0 Hz, H-5), 6.96 (1H, d, *J* = 9.0 Hz, H-6), 7.29 (1H, t, *J* = 7.5 Hz, -*para*), 7.37 (2H, t, *J* = 7.5Hz, -*meta*), 7.48 (2H, d, *J* = 7.5 Hz, -*ortho*).

¹³C NMR (CDCl₃, 150MHz) δ (ppm): 55.8, 67.1, 71.1, 114.2, 115.4, 116.4, 126.8, 127.8, 128.6, 136.5, 136.8, 147.7, 151.0, 168.8.

(5*S*, 6*R*)-5,6-Dihydro-5-(methoxymethoxy)-6-methylpyran-2-one (*ent*-1.7)
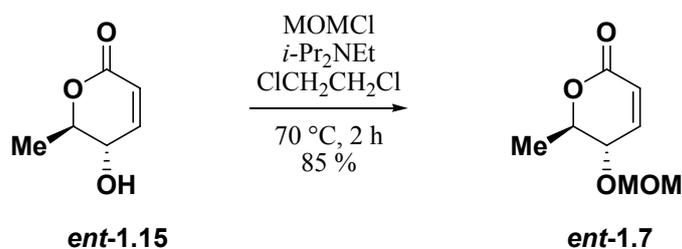
To a solution of the compound *ent*-**1.15** 5.03 g (39.3 mmol) in 1,2-dichloroethane 100 ml were added diisopropylethylamine 41 ml (235 mmol) and chloromethyl methyl ether 13 ml (171 mmol) at room

temperature. The reaction mixture was heated at 70 °C for 2 hours. After the addition of methanol 13 ml (321 mmol), the reaction mixture was evaporated in vacuo. To the residue was added H_2O 80 ml, and the mixture was extracted with ethyl acetate (3×200 ml). After evaporation in vacuo, the crude residue was purified by flash column chromatography (CH_2Cl_2-acetone 35:1) to yield the compound **ent-1.7** as a colorless syrup (5.73 g, 33.3 mmol, 85 %).

R_f: 0.41 (*n*-hexane-EtOAc 1:1)

$[\alpha]_D^{25}$ +70 ° (*c* 0.96, $CHCl_3$).

^{1}H NMR ($CDCl_3$, 600MHz) δ (ppm): 1.46 (3H, d, J = 6.5 Hz, H-7), 3.43 (3H, s, -O-CH_2-O-C*H₃*), 4.12 (1H, ddd, J = 8.5, 2.5 & 1.8 Hz, H-3), 4.47 (1H, dq, 8.5 & 6.5 Hz, H-2), 4.73 (1H, d, J = 7.0 Hz, -O-C*H₂*-O-CH_3), 4.78 (1H, d, J = 7.0 Hz, -O-C*H₂*-O-CH_3), 6.01 (1H, dd, J = 11.0 & 1.8 Hz, H-5), 6.87 (1H, dd, J = 11.0 & 2.5 Hz, H-4).

^{13}C NMR ($CDCl_3$, 150MHz) δ (ppm): 18.3, 56.0, 72.7, 77.3, 96.4, 121.0, 146.3, 162.8.

(5*R*, 6*S*)-5,6-Dihydro-5-(methoxymethoxy)-6-methylpyran-2-one (1.7)
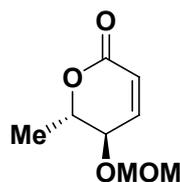
1.7

R_f: 0.41 (*n*-hexane-EtOAc 1:1)

$[\alpha]_D^{26}$ -67 ° (*c* 1.02, $CHCl_3$).

^{1}H NMR ($CDCl_3$, 600MHz) δ (ppm): 1.46 (3H, d, J = 6.5 Hz, H-7), 3.43 (3H, s, -O-CH_2-O-C*H₃*), 4.12 (1H, ddd, J = 8.5, 2.5 & 1.8 Hz, H-3), 4.47 (1H, dq, 8.5 & 6.5 Hz, H-2), 4.73 (1H, d, J = 7.0 Hz, -O-C*H₂*-O-CH_3), 4.78 (1H, d, J = 7.0 Hz, -O-C*H₂*-O-CH_3), 6.01 (1H, dd, J = 11.0 & 1.8 Hz, H-5), 6.87 (1H, dd, J = 11.0 & 2.5 Hz, H-4).

^{13}C NMR ($CDCl_3$, 150MHz) δ (ppm): 18.3, 56.0, 72.7, 77.3, 96.4, 121.0, 146.3, 162.8.

(3*R*, 4*S*)-9,10-Bis(benzyloxy)-3,4-dihydro-6-methoxy-4-(methoxymethoxy)-3-methylbenzo[*g*]isochromen-2-one (*ent*-1.6)
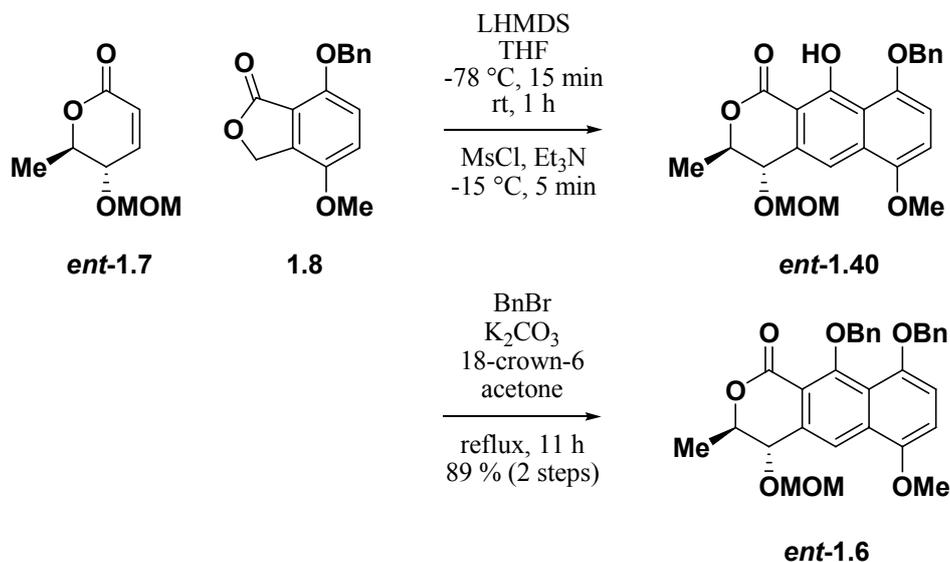
To a solution of the Michael donor **1.8** 1.86 g (6.88 mmol) in THF 74 ml was added LHMDS 1.0 M solution in THF 7.6 ml (7.6 mmol) at -78 °C. The mixture was stirred for 10 minutes, and the Michael acceptor **ent-1.7** 1.41 g (8.18 mmol) in THF 5.6 ml was added through a cannula to the mixture. After stirring for 15 minutes, LHMDS 1.0 M solution in THF 7.6 ml (7.6 mmol) was added again, and the reaction mixture was stirred at room temperature for 1 hour. Then, Et$_3$N 9.4 ml (68.8 mmol) and MsCl 1.6 ml (20.1 mmol) were added at -15 °C. After 5 minutes, H$_2$O 16 ml was added, and the mixture was evaporated in vacuo. H$_2$O 80 ml was added, and the mixture was extracted with EtOAc (3×150 ml), followed by drying with anhydrous Na$_2$SO$_4$. After filtration, the filtrate was evaporated in vacuo, and pumped up to yield the tricyclic **ent-1.40**.

To a solution of crude **ent-1.40** in acetone 110 ml were added 18-crown-6-ether 2.08 g (7.87 mmol), potassium carbonate 1.96 g (14.2 mmol), and benzyl bromide 1.23 ml (10.3 mmol). After reflux for 11 hours, the reaction mixture was evaporated in vacuo. H$_2$O 80 ml was added, and the mixture was extracted with EtOAc (3×160 ml). After evaporation in vacuo, the residue was purified by flash column chromatography (toluene-EtOAc 6:1) to yield **ent-1.6** as a pale yellow powder (3.16 g, 6.13 mmol, 89 %)

R_f: 0.31 (toluene-EtOAc 6:1)

HR-MS (FAB+): calcd. for C$_{31}$H$_{31}$O$_7$: 515.2070, found 515.2084 [M+H]$^+$.

[α]$_D$^{25} +76 ° (*c* 0.94,CHCl$_3$).

mp : 113.0-115.0 °C

IR (cm^{-1}, KBr): 2937, 1728, 1455, 1339, 1221, 1077, 1027, 738, 697.

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 1.18 (3H, d, *J* = 6.8 Hz, H-11), 3.47 (3H, s, -O-CH$_2$-O-C*H$_3$*), 3.96 (3H, s, -OMe), 4.64 (1H, d, *J* = 3.5 Hz, H-4), 4.71 (1H, d, *J* = 7.0 Hz, -C*H$_2$*Ph), 4.79 (1H, d, *J* = 7.0 Hz, -C*H$_2$*Ph), 4.80 (1H, qd, *J* = 6.8 & 3.5 Hz, H-3), 5.09 (1H, d, *J* = 12.5 Hz, -C*H$_2$*Ph), 5.12 (1H, d, *J* = 12.5

Hz, -CH_ZPh), 5.13 (1H, d, J = 10.0 Hz, -CH_ZPh), 5.28 (1H, d, J = 10.0 Hz, -CH_ZPh), 6.84 (1H, d, J = 8.5 Hz, H-8), 6.91 (1H, d, J = 8.5 Hz, H-7), 7.21-7.24 (3H, m, -CH$_2$-*Ph*), 7.24-7.27 (3H, m, -CH$_2$-*Ph*), 7.33-7.36 (2H, m, -CH$_2$-*Ph*), 7.45-7.48 (2H, m, -CH$_2$-*Ph*), 8.05 (1H, s, H-5).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): 17.9, 55.8, 55.9, 73.1, 73.6, 76.9, 78.1, 94.3, 107.2, 112.0, 116.1, 118.1, 122.9, 127.6, 127.7, 128.1, 128.4, 129.0, 130.0, 132.5, 137.0, 149.8, 150.1, 159.1, 160.7.

(3*S*, 4*R*)-9,10-Bis(benzyloxy)-3,4-dihydro-6-methoxy-4-(methoxymethoxy)-3-methylbenzo[*g*]isochromen-2-one (**1.6**)
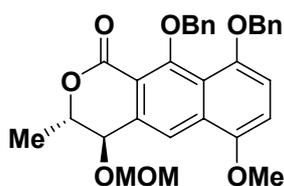
1.6

R$_f$: 0.31 (toluene-EtOAc 6:1)

[α]$_D^{25}$ -74 ° (*c* 1.03,CHCl$_3$).

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 1.18 (3H, d, J = 6.8 Hz, H-11), 3.47 (3H, s, -O-CH$_2$-O-CH_3), 3.96 (3H, s, -OMe), 4.64 (1H, d, J = 3.5 Hz, H-4), 4.71 (1H, d, J = 7.0 Hz, -CH_ZPh), 4.79 (1H, d, J = 7.0 Hz, -CH_ZPh), 4.80 (1H, qd, J = 6.8 & 3.5 Hz, H-3), 5.09 (1H, d, J = 12.5 Hz, -CH_ZPh), 5.12 (1H, d, J = 12.5 Hz, -CH_ZPh), 5.13 (1H, d, J = 10.0 Hz, -CH_ZPh), 5.28 (1H, d, J = 10.0 Hz, -CH_ZPh), 6.84 (1H, d, J = 8.5 Hz, H-8), 6.91 (1H, d, J = 8.5 Hz, H-7), 7.21-7.24 (3H, m, -CH$_2$-*Ph*), 7.24-7.27 (3H, m, -CH$_2$-*Ph*), 7.33-7.36 (2H, m, -CH$_2$-*Ph*), 7.45-7.48 (2H, m, -CH$_2$-*Ph*), 8.05 (1H, s, H-5).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): 17.9, 55.8, 55.9, 73.1, 73.6, 76.9, 78.1, 94.3, 107.2, 112.0, 116.1, 118.1, 122.9, 127.6, 127.7, 128.1, 128.4, 129.0, 130.0, 132.5, 137.0, 149.8, 150.1, 159.1, 160.7.

(1*R*, 3*R*, 4*S*)-9,10-Bis(benzyloxy)-3,4-dihydro-6-methoxy-4-(methoxymethoxy)-
3-methyl-1-propyl-1*H*-benzo[*g*]isochromene (*ent*-1.41)
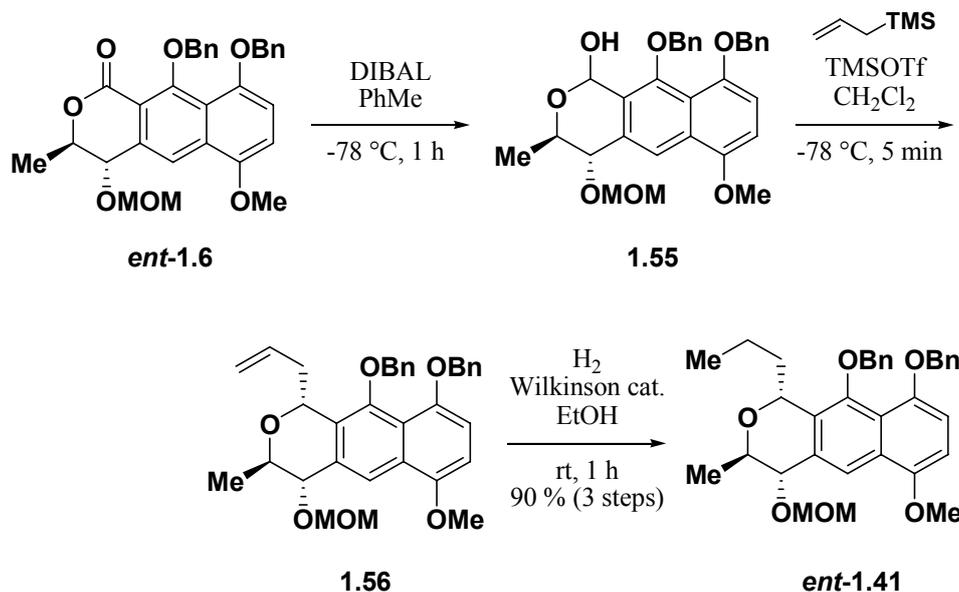
To a solution of the compound *ent*-1.6 5.72 g (11.1 mmol) in toluene 240 ml was added DIBAL 1.01 M solution in toluene 33.5 ml (33.2 mmol) at -78 °C. The reaction mixture was stirred for an hour, and MeOH 6.5 ml was added at the same temperature. After filtration with celite and EtOAc, the mixture was concentrated under reduced pressure, and pumped up to dryness to yield the lactol **1.55**.

To a solution of the lactol **1.55** in CH_2Cl_2 240 ml were added allyltrimethylsilane 35.6 ml (221 mmol) and TMSOTf 0.96 ml (5.53 mmol) at -78 °C. After 5 minutes, saturated $NaHCO_3$ 10ml was added, and heated to room temperature. H_2O 70 ml was added, and the mixture was extracted with CH_2Cl_2 (3×200 ml). After evaporation in vacuo, the residue was pumped up to dryness to yield **1.56**.

To a solution of **1.56** in EtOH 115 ml was added Wilkinson catalyst 1.18 g. The reaction mixture was stirred vigorously for an hour under hydrogen atmosphere. After evaporation in vacuo, the residue was purified by flash column chromatography (*n*-hexane-EtOAc 3:1) to yield *ent*-1.41 as pale yellow oil (4.85 g, 9.02 mmol, 89.6 %).

R_f: 0.44 (*n*-hexane-EtOAc 3:1)

HR-MS (FAB+): calcd. for $C_{34}H_{38}O_6$: 542.2668, found 542.2663 [M+H]$^+$.

$[\alpha]_D^{28}$ -79 ° (*c* 0.73,$CHCl_3$).

IR (cm^{-1}, KBr): 2956, 2932, 1600, 1455, 1339, 1260, 1062, 1029, 737, 697.

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 0.88 (3H, t, *J* = 7.3Hz, H-13), 1.29 (3H, d, *J* = 6.5 Hz, H-14), 1.36-1.46 (1H, m, H-12), 1.46-1.55 (1H, m, H-12), 1.91-1.99 (1H, m, H-11), 1.99-2.06 (1H, m, H-11), 3.57 (3H, s, -O-CH$_2$-O-C*H$_3$*), 3.95 (3H, s, -O*Me*), 4.13 (1H, dq, *J* = 7.0 & 6.5 Hz, H-3), 4.52 (1H, d, *J* = 7.0 Hz, H-4), 4.66 (1H, d, *J* = 11.3 Hz, -O-C*H$_2$*Ph), 4.90 (1H, d, *J* = 11.7 Hz, -O-C*H$_2$*O-CH$_3$), 4.92 (1H, d, *J* = 11.7 Hz, -O-C*H$_2$*O-CH$_3$), 4.96 (1H, dd, *J* = 10.0 & 2.9 Hz, H-1), 5.01 (1H, d, *J* = 11.8 Hz, -O-C*H$_2$*Ph), 5.19 (1H, d, *J* = 11.8 Hz, -O-C*H$_2$*Ph), 5.28 (1H, d, *J* = 11. 3 Hz, -O-C*H$_2$*Ph), 6.65 (1H, d, *J*

= 8.5 Hz, H-7), 6.83 (1H, d, J = 8.5 Hz, H-8), 7.23-7.27 (5H, m, -O-CH$_2$-*Ph*), 7.36-7.40 (2H, m, -O-CH$_2$-*Ph*), 8.18 (1H, s, H-5).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): 13.8, 18.6, 19.4, 35.7, 55.5, 55.7, 56.2, 67.6, 71.7, 73.1, 77.2, 78.0, 96.6, 103.2, 110.3, 118.3, 121.0, 127.5, 127.58, 127.61, 127.7, 128.2, 128.3, 131.4, 132.8, 137.3, 137.9, 148.3, 150.1, 150.4.

(1*S*, 3*S*, 4*R*)-9,10-Bis(benzyloxy)-3,4-dihydro-6-methoxy-4-(methoxymethoxy)-3-methyl-1-propyl-1*H*-benzo[*g*]isochromene (1.41)
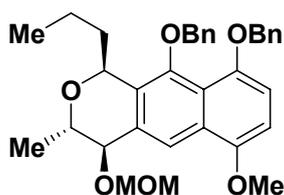
1.41

R_f: 0.44 (*n*-hexane-EtOAc 3:1)

[α]$_D^{25}$ +78 ° (*c* 0.97, CHCl$_3$).

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 0.88 (3H, t, J = 7.3Hz, H-13), 1.29 (3H, d, J = 6.5 Hz, H-14), 1.36-1.46 (1H, m, H-12), 1.46-1.55 (1H, m, H-12), 1.91-1.99 (1H, m, H-11), 1.99-2.06 (1H, m, H-11), 3.57 (3H, s, -O-CH$_2$-O-C*H$_3$*), 3.95 (3H, s, -O*Me*), 4.13 (1H, dq, J = 7.0 & 6.5 Hz, H-3), 4.52 (1H, d, J = 7.0 Hz, H-4), 4.66 (1H, d, J = 11.3 Hz, -O-C*H$_2$*-Ph), 4.90 (1H, d, J = 11.7 Hz, -O-C*H$_2$*-O-CH$_3$), 4.92 (1H, d, J = 11.7 Hz, -O-C*H$_2$*-O-CH$_3$), 4.96 (1H, dd, J = 10.0 & 2.9 Hz, H-1), 5.01 (1H, d, J = 11.8 Hz, -O-C*H$_2$*Ph), 5.19 (1H, d, J = 11.8 Hz, -O-C*H$_2$*Ph), 5.28 (1H, d, J = 11. 3 Hz, -O-C*H$_2$*Ph), 6.65 (1H, d, J = 8.5 Hz, H-7), 6.83 (1H, d, J = 8.5 Hz, H-8), 7.23-7.27 (5H, m, -O-CH$_2$-*Ph*), 7.36-7.40 (2H, m, -O-CH$_2$-*Ph*), 8.18 (1H, s, H-5).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): 13.8, 18.6, 19.4, 35.7, 55.5, 55.7, 56.2, 67.6, 71.7, 73.1, 77.2, 78.0, 96.6, 103.2, 110.3, 118.3, 121.0, 127.5, 127.58, 127.61, 127.7, 128.2, 128.3, 131.4, 132.8, 137.3, 137.9, 148.3, 150.1, 150.4.

(1*R*, 3*R*, 4*S*)-9,10-Bis(benzyloxy)-3,4-dihydro-4-hydroxy-6-methoxy-3-methyl-1-propyl-1*H*-benzo[*g*]isochromene-8-carbaldehyde (1.57)
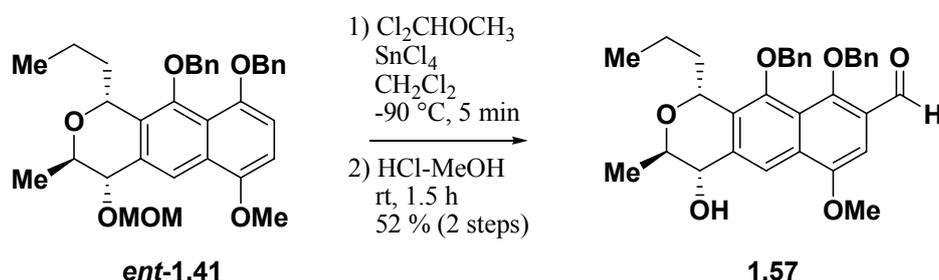

To a solution of $SnCl_4$ 1.1 ml (9.40 mmol) in CH_2Cl_2 74 ml was added dichloromethyl methyl ether 0.82 ml (9.07 mmol) at -70 °C. After 10 minutes, the compound ***ent*-1.41** 2.47 g (4.54 mmol) in CH_2Cl_2 74 ml was added to the mixture through a cannula at -90 °C. After 5 minutes, Et_3N 35 ml (257 mmol) was added, the mixture was evaporated under reduced pressure, and pumped up to dryness.

To the residue was added HCl-MeOH 50 ml, and the mixture was stirred for 1.5 hours. The reaction mixture was directly purified by flash column chromatography (toluene-EtOAc 10:1) to remove HCl-MeOH. After collection and evaporation of the desired fractions, the residue was purified again by flash column chromatography (*n*-hexane-EtOAc 3:1) to yield **1.57** as a orange solid (1.25 g, 2.37 mmol, 52 %).

R_f: 0.40 (*n*-hexane-EtOAc 2:1)

HR-MS (FAB+): calcd. for $C_{33}H_{35}O_6$: 527.2434, found 527.2411 $[M+H]^+$.

$[\alpha]_D^{26}$ -218 ° (*c* 0.98, $CHCl_3$)

mp : 57.2-57.8 °C

IR (cm^{-1}, KBr): 3435, 2958, 2932, 2871, 1675, 1397, 1357, 1343, 754, 697.

^{1}H NMR ($CDCl_3$, 600MHz) δ (ppm): 0.96 (3H, t, *J* = 7.5 Hz, H-13), 1.37 (3H, d, *J* = 6.0 Hz, H-14), 1.45-1.63 (2H, m, H-12), 1.94-2.02 (1H, m, H-11), 2.03-2.10 (1H, m, H-11), 2.12-2.19 (1H, m, -OH), 3.91 (1H, dq, 8.3 & 6.0 Hz, H-3), 4.04 (3H, s, -OMe), 4.52 (1H, dd, 8.3 & 8.3 Hz, H-4), 4.71 (1H, d, *J* = 10.5 Hz, -C*H$_2$*Ph), 4.72 (1H, d, *J* = 11.0 Hz, -C*H$_2$*Ph), 5.00 (1H, dd, *J* = 10.5 & 3.0 Hz, H-1), 5.26 (1H, d, *J* = 11.0 Hz, -*CH$_2$*Ph), 5.27 (1H, d, *J* = 10.5 Hz, -C*H$_2$*Ph), 7.08 (1H, s, H-7), 7.13-7.16 (2H, m, -CH$_2$-*Ph*), 7.24-7.28 (2H, m, -C*H$_2$*Ph), 7.28-7.32 (1H, m, -C*H$_2$*Ph), 7.32-7.37 (5H, m, -CH$_2$-*Ph*), 8.35 (1H, s, H-5), 10.21 (1H, s, -CHO).

^{13}C NMR ($CDCl_3$, 150MHz) δ (ppm): 13.8, 18.4, 19.4, 35.2, 55.8, 68.3, 71.6, 72.1, 77.8, 80.0, 98.5, 98.6, 117.4, 121.7, 126.8, 128.07, 128.12, 128.4, 128.5, 128.6, 129.3, 131.4, 132.5, 135.4, 137.1, 139.3, 150.8, 152.2, 153.8, 190.2.

(1*S*, 3*S*, 4*R*)-9,10-Bis(benzyloxy)-3,4-dihydro-4-hydroxy-6-methoxy-3-methyl-1-propyl-1*H*-benzo[*g*]isochromene-8-carbaldehyde (*ent*-1.57)
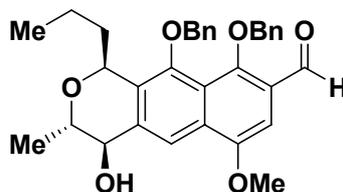
ent-**1.57**

R_f: 0.40 (*n*-hexane-EtOAc 2:1)

$[\alpha]_D^{25}$ +217 ° (*c* 0.98, CHCl$_3$)

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 0.96 (3H, t, *J* = 7.5 Hz, H-13), 1.37 (3H, d, *J* = 6.0 Hz, H-14), 1.45-1.63 (2H, m, H-12), 1.94-2.02 (1H, m, H-11), 2.03-2.10 (1H, m, H-11), 2.12-2.19 (1H, m, -OH), 3.91 (1H, dq, 8.3 & 6.0 Hz, H-3), 4.04 (3H, s, -OMe), 4.52 (1H, dd, 8.3 & 8.3 Hz, H-4), 4.71 (1H, d, *J* = 10.5 Hz, -C*H*$_2$-Ph), 4.72 (1H, d, *J* = 11.0 Hz, -C*H*$_2$-Ph), 5.00 (1H, dd, *J* = 10.5 & 3.0 Hz, H-1), 5.26 (1H, d, *J* = 11.0 Hz, -*CH*$_2$-Ph), 5.27 (1H, d, *J* = 10.5 Hz, -C*H*$_2$-Ph), 7.08 (1H, s, H-7), 7.13-7.16 (2H, m, -CH$_2$-*Ph*), 7.24-7.28 (2H, m, -C*H*$_2$-Ph), 7.28-7.32 (1H, m, -C*H*$_2$-Ph), 7.32-7.37 (5H, m, -CH$_2$-*Ph*), 8.35 (1H, s, H-5), 10.21 (1H, s, -CHO).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): 13.8, 18.4, 19.4, 35.2, 55.8, 68.3, 71.6, 72.1, 77.8, 80.0, 98.5, 98.6, 117.4, 121.7, 126.8, 128.07, 128.12, 128.4, 128.5, 128.6, 129.3, 131.4, 132.5, 135.4, 137.1, 139.3, 150.8, 152.2, 153.8, 190.2.

(*R*)-9,10-Bis(benzyloxy)-6-methoxy-3-methyl-1-propyl-1*H*-benzo[*g*]isochromene-8-carbaldehyde (*ent*-1.42)

1.57   Burgess salt / THF / 50 °C, 1.5 h / 71 %   *ent*-**1.42**
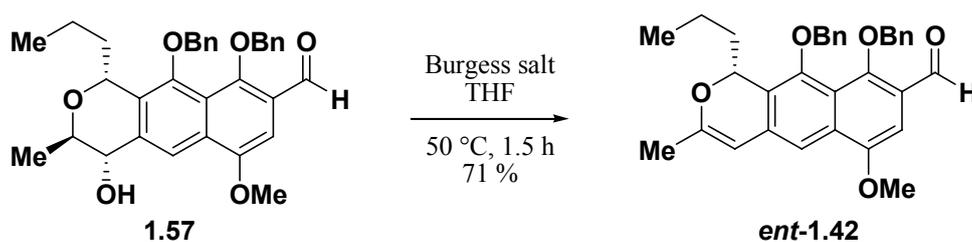
To a solution of the aldehyde **1.57** 0.569 g (1.08 mmol) in THF 28 ml was added Burgess salt 0.408 g (1.71 mmol) at room temperature. After stirring at 50 °C for 1.5 hours, the reaction mixture was evaporated in vacuo. The residue was purified by flash column chromatography (*n*-hexane-EtOAc 8:1) to yield *ent*-**1.42** as orange oil (0.388 g, 0.763 mmol, 71 %).

R_f: 0.40 (*n*-hexane-EtOAc 8:1)

HR-MS (FAB+): calcd. for C$_{33}$H$_{32}$O$_5$: 508.2250, found 508.2260 [M+H]$^+$.

$[\alpha]_D^{26}$ +157 ° (*c* 1.01, CHCl$_3$)

IR (cm^{-1}, KBr): 2958, 2871, 1673, 1610, 1397, 1353, 1199, 1051, 753, 699.

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 0.92 (3H, t, J = 7.3 Hz, H-13), 1.40-1.49 (1H, m,H-12), 1.49-1.61 (2H, m, H-11 & H-12), 1.98 (3H, d, J = 1.0 Hz, H-14), 2.04-2.12 (1H, m, H-11), 4.01 (3H, s, -OMe), 4.79 (1H, d, J = 11.0 Hz, -C$H_\mathcal{2}$Ph), 4.90 (1H, d, J = 11.0 Hz, -C$H_\mathcal{2}$Ph), 5.17 (1H, d, J = 11.0 Hz, -C$H_\mathcal{2}$Ph), 5.19 (1H, d, J = 11.0 Hz, -C$H_\mathcal{2}$Ph), 5.66 (1H, dd, J = 9.8 & 4.0 Hz, H-1), 5.77 (1H, d, J = 1.0 Hz, H-4), 7.07 (1H, s, H-7), 7.15-7.17 (2H, m, -CH$_2$-Ph), 7.23-7.26 (2H, m, -CH$_2$-Ph), 7.27-7.31 (1H, m, -CH$_2$-Ph), 7.32-7.38 (3H, m, -CH$_2$-Ph), 7.39-7.42 (2H, m, -CH$_2$-Ph), 7.61 (1H, s, H-5), 10.20 (1H, s, -CHO).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): 13.8, 18.7, 20.3, 36.4, 55.8, 73.8, 77.7, 80.2, 99.02, 99.05, 100.48, 100.50, 111.5, 120.9, 125.0, 125.4, 127.9, 128.42, 128.44, 129.2, 132.6, 133.2, 135.6, 137.2, 150.1, 151.9, 154.4, 154.9, 190.1.

(*S*)-9,10-Bis(benzyloxy)-6-methoxy-3-methyl-1-propyl-1*H*-benzo[*g*]isochromene-8-carbaldehyde (**1.42**)
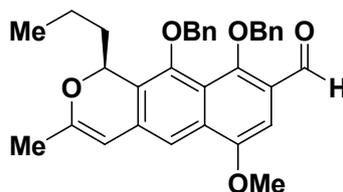
1.42

R_f: 0.40 (*n*-hexane-EtOAc 8:1)

[α]$_D{}^{25}$ -157 ° (*c* 1.05, CHCl$_3$)

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 0.92 (3H, t, J = 7.3 Hz, H-13), 1.40-1.48 (1H, m,H-12), 1.49-1.61 (2H, m, H-11 & H-12), 1.98 (3H, d, J = 1.0 Hz, H-14), 2.04-2.12 (1H, m, H-11), 4.01 (3H, s, -OMe), 4.79 (1H, d, J = 11.0 Hz, -C$H_\mathcal{2}$Ph), 4.90 (1H, d, J = 11.0 Hz, -C$H_\mathcal{2}$Ph), 5.17 (1H, d, J = 11.0 Hz, -C$H_\mathcal{2}$Ph), 5.19 (1H, d, J = 11.0 Hz, -C$H_\mathcal{2}$Ph), 5.66 (1H, dd, J = 9.8 & 4.0 Hz, H-1), 5.77 (1H, d, J = 1.0 Hz, H-4), 7.07 (1H, s, H-7), 7.15-7.17 (2H, m, -CH$_2$-Ph), 7.23-7.26 (2H, m, -CH$_2$-Ph), 7.27-7.31 (1H, m, -CH$_2$-Ph), 7.32-7.38 (3H, m, -CH$_2$-Ph), 7.39-7.42 (2H, m, -CH$_2$-Ph), 7.61 (1H, s, H-5), 10.20 (1H, s, -CHO).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): 13.8, 18.7, 20.3, 36.4, 55.8, 73.8, 77.7, 80.2, 99.03, 99.04, 100.49, 100.49, 111.5, 120.9, 125.0, 125.4, 127.9, 128.42, 128.45, 129.2, 132.6, 133.2, 135.6, 137.2, 150.1, 151.9, 154.5, 154.9, 190.1.

(2*R*, 3*S*)-2,4-Bis(*tert*-butyldimethylsiloxy)butane-1,3-diol (1.27)
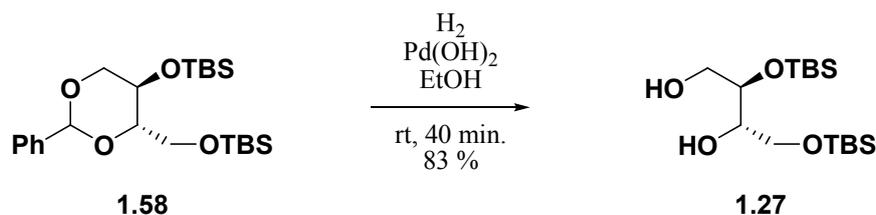
To a solution of **1.58** 0.297 g (0.676 mmol) in EtOH 8.9 ml was added Pd(OH)$_2$ 30.9 mg. After stirring vigorously for 40 minutes under hydrogen atmosphere, the reaction mixture was filtered with celite, and the filtrate was evaporated in vacuo. The residue was purified by flash column chromatography (*n*-hexane-EtOAc 5:1) to yield the diol **1.27** as colorless syrup (0.197 g, 0.561 mmol, 83 %).

R_f: 0.36 (*n*-hexane-EtOAc 3:1)

HR-MS (FAB+): calcd. for C$_{16}$H$_{39}$O$_4$Si$_2$: 351.2387, found 351.2364 [M+H]$^+$.

[α]$_D{}^{25}$ -11 ° (*c* 1.14, CHCl$_3$)

IR (cm^{-1}, KBr): 3441, 2929, 2858, 1361, 1253, 835.

^{1}H NMR (DMSO-*d*$_6$, 600MHz) δ (ppm): 0.02 (3H, s, -Si-*Me$_2$*), 0.03 (3H, s, -Si-*Me$_2$*), 0.04 (3H, s, -Si-*Me$_2$*), 0.05 (3H, s, -Si-*Me$_2$*), 0.85 (9H, s, -Si-*t-Bu*), 0.86 (9H, s, -Si-*t-Bu*), 3.32-3.41 (1H, m, H-1), 3.41-3.47 (1H, m, H-3), 3.52 (1H, dd, *J* = 10.5 & 6.0 Hz, H-4), 3.55 (1H, ddd, *J* = 11.0, 4.5 & 4.5 Hz, H-1), 3.60 (1H, ddd, *J* = 5.5, 5.5 & 4.5 Hz, H-2), 3.68 (1H, dd, *J* = 10.5 & 4.0 Hz, H-4), 4.41 (1H, dd, *J* = 5.5 & 4.5 Hz, -OH), 4.55 (1H, d, *J* = 4.8 Hz, -OH).

^{13}C NMR (DMSO-*d*$_6$, 150MHz) δ (ppm): -5.0, -4.7, -4.0, 18.2, 18.3, 26.1, 63.4, 64.6, 72.5, 74.9.

(2*S*, 3*R*)-2,4-Bis(*tert*-butyldimethylsiloxy)butane-1,3-diol (*ent*-1.27)

To a solution of **1.59** 4.25 g (8.01 mmol) in EtOH 128 ml was added Pd(OH)$_2$ 462 mg. After stirring vigorously for 1.5 hours under hydrogen atmosphere, the reaction mixture was filtered with celite, and the filtrate was evaporated in vacuo. The residue was purified by flash column chromatography (*n*-hexane-toluene 3:2) to yield the diol **ent-1.27** as colorless syrup (2.38 g, 6.79 mmol, 85 %).
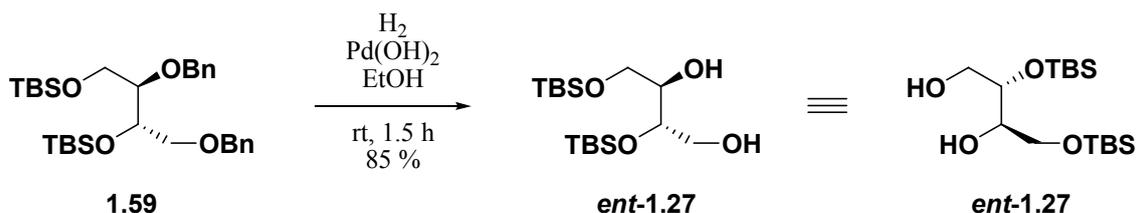
R_f: 0.36 (*n*-hexane-EtOAc 3:1)

[α]$_D{}^{25}$ +12 ° (*c* 1.15, CHCl$_3$)

^{1}H NMR (DMSO-*d*$_6$, 600MHz) δ (ppm): 0.02 (3H, s, -Si-*Me$_2$*), 0.03 (3H, s, -Si-*Me$_2$*), 0.04 (3H, s, -Si-*Me$_2$*), 0.05 (3H, s, -Si-*Me$_2$*), 0.85 (9H, s, -Si-*t-Bu*), 0.86 (9H, s, -Si-*t-Bu*), 3.32-3.41 (1H, m, H-1),

3.41-3.47 (1H, m, H-3), 3.52 (1H, dd, J = 10.5 & 6.0 Hz, H-4), 3.55 (1H, ddd, J = 11.0, 4.5 & 4.5 Hz, H-1), 3.60 (1H, ddd, J = 5.5, 5.5 & 4.5 Hz, H-2), 3.68 (1H, dd, J = 10.5 & 4.0 Hz, H-4), 4.41 (1H, dd, J = 5.5 & 4.5 Hz, -OH), 4.55 (1H, d, J = 4.8 Hz, -OH).

^{13}C NMR (DMSO-d_6, 150MHz) δ (ppm): -5.0, -4.7, -4.0, 18.2, 18.3, 26.1, 63.4, 64.6, 72.5, 74.9.

(2R, 4S, 5R)-2-((R)-9,10-Bis(benzyloxy)-6-methoxy-3-methyl-1-propyl-1H-benzo[g]isochromen-8-yl)-4-(*tert*-butyldimethylsiloxymethyl)-5-(*tert*-butyldimethylsiloxy)-1,3-dioxane ((1R, 5'S)-1.51)

ent-1.42   **(1R, 5'S)-1.51**
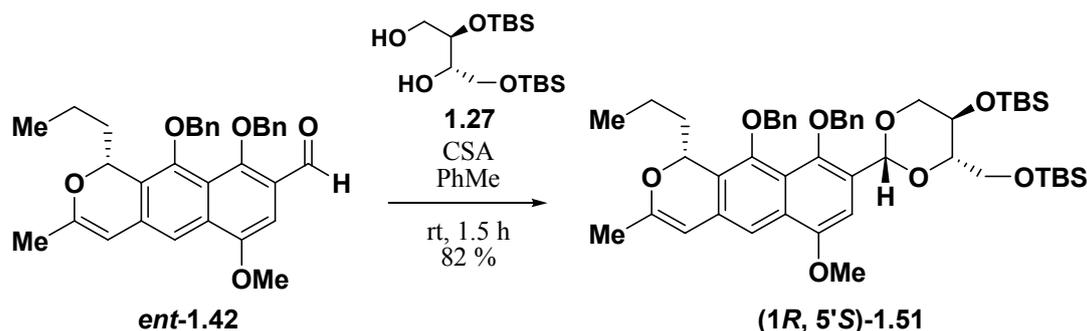
To a solution of **ent-1.42** 0.388 g (0.763 mmol) and CSA 0.187 g (0.805 mmol) in toluene 19 ml was added a solution of the diol **1.27** 0.409 g (1.17 mmol) in toluene 12 ml through a cannula. After 1.5 hours, Et$_3$N 1.43 ml (10.5 mmol) was added at 0 °C. The mixture was evaporated in vacuo, and the residue was purified by flash column chromatography (*n*-hexane-toluene 1:3) to yield **(1R, 5'S)-1.51** as a white solid (0.525 g, 0.624 mmol, 82 %).

R_f: 0.40 (*n*-hexane-toluene 1:1)

HR-MS (FAB+): calcd. for C$_{49}$H$_{69}$O$_8$Si$_2$: 841.4531, found 841.4553 [M+H]$^+$.

mp: 60.5-61.5 °C

$[\alpha]_D^{26}$ +52 ° (*c* 1.04, CHCl$_3$)

IR (cm^{-1}, KBr): 2955, 2929, 2856, 1404, 1359, 1253, 1205, 1103, 1055, 836.

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 0.07 (3H, s, -Si-Me_2), 0.11 (3H, s, -Si-Me_2), 0.12 (3H, s, -Si-Me_2), 0.14 (3H, s, -Si-Me_2), 0.91 (9H, s, -Si-t-Bu), 0.91 (3H, t, J = 7.3 Hz, H-13), 0.93 (9H, s, -Si-t-Bu), 1.38-1.47 (1H, m, H-12), 1.49-1.60 (2H, m, H-11 & H-12), 1.96 (3H, s, H-14), 2.01-2.10 (1H, m, H-11), 3.40 (1H, dd, J = 10.5 & 10.5Hz, H-3'), 3.54 (1H, ddd, J = 9.0, 3.5 & 1.5 Hz, H-5'), 3.86 (1H, dd, J = 11.8 & 1.5 Hz, H-6'), 3.93 (1H, dd, J = 11.8 & 3.5 Hz, H-6'), 3.98-4.04 (1H, m, H-4'), 4.01 (3H, s, -OMe), 4.10 (1H, dd, J = 10.5 & 5.5 Hz, H-3'), 4.71 (1H, d, J = 10.5 Hz, -CH_2Ph), 4.83 (1H, d, J = 10.5 Hz, -CH_2Ph), 5.09 (1H, d, J = 10.5 Hz, -CH_2Ph), 5.18 (1H, d, J = 10.5 Hz, -CH_2Ph), 5.62 (1H, dd, J = 9.5 & 3.5 Hz, H-1), 5.74 (1H, d, J = 0.5 Hz, H-4), 6.05 (1H, s, H-1'), 7.05 (1H, s, H-7), 7.25-7.33 (10H, m, -CH$_2$-Ph), 7.60 (1H, s, H-5).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): -5.2, -4.93, -4.89, -4.4, 13.8, 17.9, 18.4, 18.7, 20.2, 25.68, 25.71, 25.9, 26.0, 36.4, 55.7, 61.6, 62.0, 71.7, 73.8, 77.5, 78.4, 83.2, 97.3, 100.49, 100.51, 101.9, 102.0, 111.0,

121.1, 124.3, 126.5, 127.5, 127.8, 128.15, 128.18, 128.5, 129.1, 130.0, 137.7, 137.8, 145.8, 149.2, 151.9, 152.9.

(2S, 4R, 5S)-2-((R)-9,10-Bis(benzyloxy)-6-methoxy-3-methyl-1-propyl-1H-benzo[g]isochromen-8-yl)-4-(*tert*-butyldimethylsiloxymethyl)-5-(*tert*-butyldimethylsiloxy)-1,3-dioxane (*ent*-1.51)
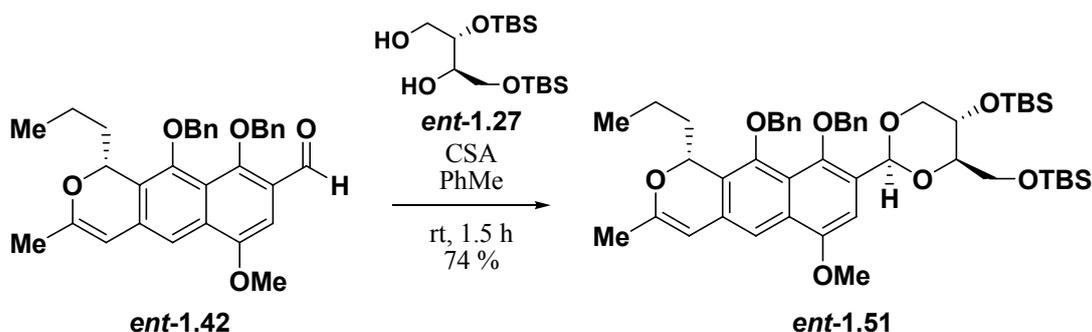
To a solution of **ent-1.42** 0.500 g (0.984 mmol) and CSA 0.253 g (1.09 mmol) in toluene 15 ml was added a solution of the diol **ent-1.27** 0.445 g (1.27 mmol) in toluene 7.5 ml through a cannula. After 1.5 hours, Et₃N 2.50 ml (18.3 mmol) was added at 0 °C. The mixture was evaporated in vacuo, and the residue was purified by flash column chromatography (*n*-hexane-toluene 1:3) to yield **ent-1.51** as a white solid (0.609 g, 0.725 mmol, 74 %).

R_f: 0.40 (*n*-hexane-toluene 1:1)
HR-MS (FAB+): calcd. for $C_{49}H_{69}O_8Si_2$: 841.4531, found 841.4507 [M+H]⁺.
mp: 58.9-59.5 °C
[α]$_D^{24}$ +36 ° (*c* 1.07, CHCl₃)
IR (cm⁻¹, KBr): 2955, 2857, 1359, 1253, 1205, 1103, 837.
¹H NMR (CDCl₃, 600MHz) δ (ppm): -0.01 (3H, s, -Si-*Me₂*), 0.04 (3H, s, -Si-*Me₂*), 0.11 (3H, s, -Si-*Me₂*), 0.12 (3H, s, -Si-*Me₂*), 0.87 (9H, s, -Si-*t-Bu*), 0.88 (3H, *J* = 5.0 Hz, H-13), 0.90 (9H, s, -Si-*t-Bu*), 1.36-1.43 (1H, m, H-12), 1.44-1.59 (2H, m, H-11 & H-12), 1.94 (3H, s, H-14), 2.00-2.07 (1H, m, H-11), 3.41 (1H, ddd, *J* = 9.3, 3.8 & 1.5 Hz, H-5'), 3.51 (1H, dd, *J* = 10.5 & 10.5 Hz, H-3'), 3.76 (1H, dd, *J* = 11.5 & 1.5 Hz, H-6'), 3.85 (1H, dd, *J* = 11.5 & 3.8 Hz, H-6'), 3.94 (1H, ddd, *J* = 10.5, 9.3 & 5.5 Hz, H-4'), 3.99 (3H, s, -OMe), 4.14 (1H, dd, *J* = 10.5 & 5.5 Hz, H-3'), 4.74 (1H, d, *J* = 10.5 Hz, -C*H₂*-Ph), 4.84 (1H, d, *J* = 11.0 Hz, -C*H₂*-Ph), 5.01 (1H, d, *J* = 10.5 Hz, -C*H₂*-Ph), 5.13 (1H, d, *J* = 11.0 Hz, -C*H₂*-Ph), 5.60 (1H, dd, *J* = 9.3 & 3.5 Hz, H-1), 5.73 (1H, s, H-4), 6.00 (1H, s, H-1'), 7.00 (1H, s, H-7), 7.22-7.28 (6H, m, -CH₂-*Ph*), 7.28-7.31 (4H, m, -CH₂-*Ph*), 7.57 (1H, s, H-5).
¹³C NMR (CDCl₃, 150MHz) δ (ppm): -5.3, -4.95, -4.89, -4.4, 13.8, 17.9, 18.4, 18.7, 20.2, 25.7, 25.7, 25.9, 25.9, 36.3, 55.7, 61.8, 62.0, 71.8, 73.8, 77.5, 78.5, 83.2, 97.2, 100.5, 102.2, 111.0, 121.1, 124.3, 126.6, 127.5, 127.6, 127.8, 128.17, 128.18, 128.6, 129.1, 130.0, 137.67, 137.74, 145.6, 149.2, 151.9, 152.9.

(2*R*, 4*S*, 5*R*)-2-((*S*)-9,10-Bis(benzyloxy)-6-methoxy-3-methyl-1-propyl-

1*H*-benzo[*g*]isochromen-8-yl)-4-(*tert*-butyldimethylsiloxymethyl)-5-(*tert*-butyldimethylsiloxy)-1,3-dioxane (**1.51**)

1.51
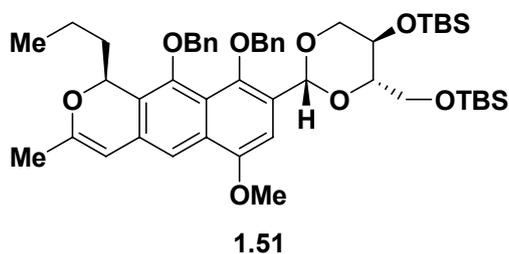
*R*f: 0.40 (*n*-hexane-toluene 1:1)

[α]$_D^{27}$ -33 ° (*c* 1.03, CHCl$_3$)

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): -0.01 (3H, s, -Si-*Me$_2$*), 0.04 (3H, s, -Si-*Me$_2$*), 0.11 (3H, s, -Si-*Me$_2$*), 0.12 (3H, s, -Si-*Me$_2$*), 0.87 (9H, s, -Si-*t*-*Bu*), 0.88 (3H, *J* = 7.3 Hz, H-13), 0.90 (9H, s, -Si-*t*-*Bu*), 1.36-1.43 (1H, m, H-12), 1.44-1.59 (2H, m, H-11 & H-12), 1.94 (3H, br s, H-14), 2.00-2.07 (1H, m, H-11), 3.41 (1H, ddd, *J* = 9.3, 3.8 & 1.5 Hz, H-5'), 3.51 (1H, dd, *J* = 10.5 & 10.5 Hz, H-3'), 3.76 (1H, dd, *J* = 11.5 & 1.5 Hz, H-6'), 3.85 (1H, dd, *J* = 11.5 & 3.8 Hz, H-6'), 3.94 (1H, ddd, *J* = 10.5, 9.3 & 5.5 Hz, H-4'), 3.99 (3H, s, -OMe), 4.14 (1H, dd, *J* = 10.5 & 5.5 Hz, H-3'), 4.74 (1H, d, *J* = 10.5 Hz, -C*H$_2$*-Ph), 4.84 (1H, d, *J* = 11.0 Hz, -C*H$_2$*-Ph), 5.01 (1H, d, *J* = 10.5 Hz, -C*H$_2$*-Ph), 5.13 (1H, d, *J* = 11.0 Hz, -C*H$_2$*-Ph), 5.60 (1H, dd, *J* = 9.3 & 3.5 Hz, H-1), 5.72 (1H, br s, H-4), 6.00 (1H, s, H-1'), 7.00 (1H, s, H-7), 7.22-7.31 (10H, m, -CH$_2$-*Ph*), 7.57 (1H, s, H-5).

^{13}C NMR (CDCl$_3$, 150MHz) δ (ppm): -5.3, -4.95, -4.89, -4.4, 13.8, 17.9, 18.4, 18.7, 20.2, 25.69, 25.72, 25.92, 25,94 36.4, 55.7, 61.8, 62.0, 71.8, 73.8, 77.5, 78.5, 83.2, 97.2, 100.5, 102.2, 111.0, 121.1, 124.3, 126.6, 127.5, 127.6, 127.8, 128.2, 128.2, 128.6, 129.1, 130.0, 137.67, 137.74, 145.6, 149.2, 151.9, 152.9.

(2*S*, 4*R*, 5*S*)-2-((*S*)-9,10-Bis(benzyloxy)-6-methoxy-3-methyl-1-propyl-

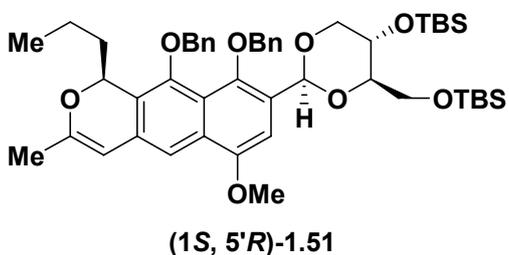1*H*-benzo[*g*]isochromen-8-yl)-4-(*tert*-butyldimethylsiloxymethyl)-5-(*tert*-butyldimethylsiloxy)-1,3-dioxane ((1*S*, 5'*R*)-**1.51**)

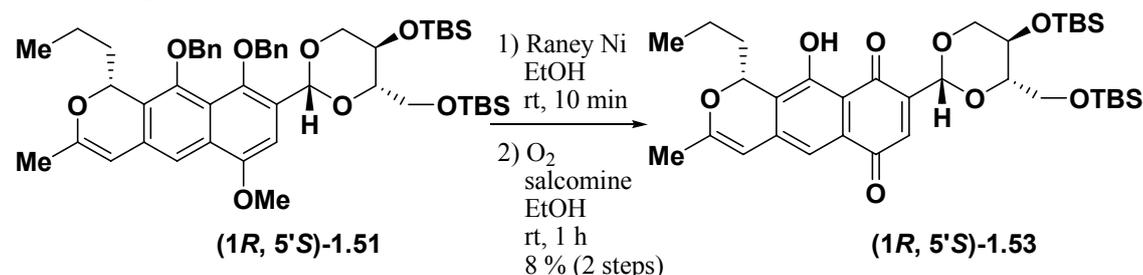(**1*S*, 5'*R***)-**1.51**

*R*f: 0.40 (*n*-hexane-toluene 1:1)

[α]$_D^{25}$ -51 ° (*c* 1.05, CHCl$_3$)

^{1}H NMR (CDCl$_3$, 600MHz) δ (ppm): 0.05 (3H, s, -Si-*Me$_2$*), 0.10 (3H, s, -Si-*Me$_2$*), 0.11 (3H, s, -Si-*Me$_2$*), 0.13 (3H, s, -Si-*Me$_2$*), 0.89 (3H, t, *J* = 7.3 Hz, H-13), 0.90 (9H, s, -Si-*t*-*Bu*), 0.92 (9H, s, -Si-*t*-*Bu*),

1.36-1.45 (1H, m, H-12), 1.47-1.58 (2H, m, H-11 & H-12), 1.94 (3H, br s, H-14), 2.00-2.07 (1H, m, H-11), 3.38 (1H, dd, J = 10.5 & 10.5 Hz, H-3'), 3.52 (1H, ddd, J = 9.0, 3.5 & 1.5 Hz, H-5'), 3.85 (1H, dd, J = 11.8 & 1.5 Hz, H-6'), 3.92 (1H, dd, J = 11.8 & 3.5 Hz, H-6'), 3.96-4.02 (1H, m, H-4'), 3.99 (3H, s, -OMe), 4.08 (1H, dd, J = 10.5 & 5.5 Hz, H-3'), 4.69 (1H, d, J = 10.5 Hz, -CH_2-Ph), 4.81 (1H, d, J = 10.5 Hz, -CH_2-Ph), 5.07 (1H, d, J = 10.5 Hz, -CH_2-Ph), 5.16 (1H, d, J = 10.5 Hz, -CH_2-Ph), 5.60 (1H, dd, J = 9.5 & 3.5 Hz, H-1), 5.73 (1H, br s, H-4), 6.03 (1H, s, H-1'), 7.03 (1H, s, H-7), 7.24-7.28 (6H, m, -CH_2-Ph), 7.28-7.32 (4H, m, -CH_2-Ph), 7.58 (1H, s, H-5).

^{13}C NMR (CDCl_3, 150MHz) δ (ppm): -5.2, -4.93, -4.90, -4.4, 13.8, 17.9, 18.4, 18.7, 20.2, 25.7, 25.7, 26.0, 26.0, 36.4, 55.7, 61.6, 62.0, 71.8, 73.9, 77.5, 78.4, 83.2, 97.3, 100.5, 100.5, 101.9, 102.0, 102.0, 111.0, 121.1, 124.3, 126.5, 127.53, 127.54, 127.8, 128.15, 128.18, 128.5, 129.1, 130.0, 137.7, 137.8, 145.8, 149.2, 151.9, 152.9.

(*R*)-10-hydroxy-8-((2*R*, 4*S*, 5*R*)- 5-(*tert*-butyldimethylsiloxy)-4-(*tert*-butyldimethylsiloxymethyl)-1,3-dioxan-2-yl)- 3-methyl-1-propyl-1*H*-benzo[*g*]isochromene-6,9-dione ((1*R*, 5'*S*)-1.53)

(1R, 5'S)-1.51 → **(1R, 5'S)-1.53**

1) Raney Ni EtOH rt, 10 min
2) O_2 salcomine EtOH rt, 1 h 8 % (2 steps)

To a solution of **(1R, 5'S)-1.51** 150.3 mg (0.179 mmol) in EtOH 15 ml was added Raney Ni (W4) 2.7 g, and the mixture was stirred for 10 minutes. The mixture was filtrated and washed with EtOH. Salcomine 24.0 mg (0.0738 mmol) was added to the mixture, which was vigorously stirred for an hour under oxygen atmosphere. The reaction mixture was evaporated in vacuo, and the residue was purified by subsequent column chromatography (toluene), three preparative TLC (*n*-hexane-tert-butyl methyl ether 10 : 1), and three column chromatography (*n*-hexane-toluene 1 :1) to give the naphthoquinone **(1R, 5'S)-1.53** as red solids (8.9 mg, 0.0138 mmol, 8 %).

R_f: 0.40 (*n*-hexane-toluene 1 : 1)

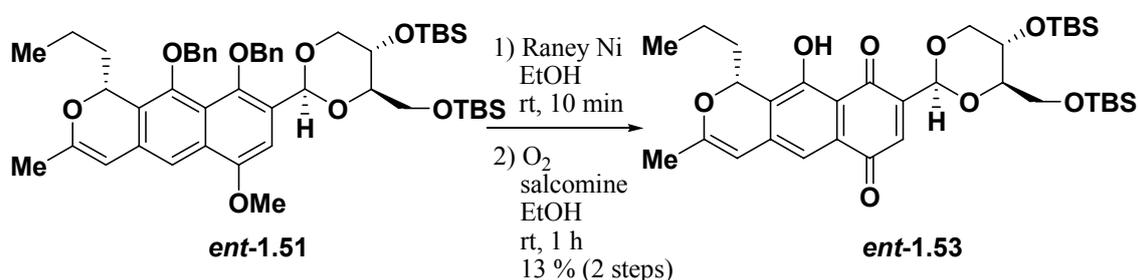HR-MS (FAB+): calcd. for $C_{34}H_{53}O_8Si_2$: 645.3279; found 645.3250.

mp: 123.0-124.5 °C

IR (cm^{-1}, KBr): 2956, 2930, 2857, 1650, 1602, 1387, 1296, 1253, 1108, 837, 778.

^{1}H NMR (C_6D_6, 600 MHz) δ (ppm): -0.04 (3H, s, -Si-Me_2), 0.06 (3H, s, -Si-Me_2), 0.11 (3H, s, -Si-Me_2), 0.14 (3H, s, -Si-Me_2), 0.90 (3H, t, J = 7.0 Hz, H-13), 0.91 (9H, s, -Si-*t-Bu*), 1.00 (9H, s, -Si-*t-Bu*), 1.40-1.49 (1H, m, H-12), 1.51-1.59 (2H, m, H-11 & H-12), 1.60 (3H, br s, H-14), 1.94-2.02 (1H, m, H-11), 3.42 (1H, dd, J = 10.5 & 10.5 Hz, H-3'), 3.44 (1H, ddd, J = 9.5, 3.5 & 1.8 Hz, H-5'), 3.83 (1H, dd,

J = 11.5 & 1.8 Hz, H-6'), 3.89 (1H, dd, J = 11.5 & 3.5 Hz, H-6'), 3.95 (1H, ddd, J = 10.5, 9.5 & 5.5 Hz, H-4'), 4.09 (1H, dd, J = 10.5 & 5.5 Hz, H-3'), 5.14 (1H, br s, H-4), 5.69 (1H, s, H-1'), 5.78 (1H, dd, J = 9.5 & 3.5 Hz, H-1), 7.19 (1H, s, H-5), 7.37 (1H, s, H-7), 12.60 (1H, s, -OH).

^{13}C NMR (C$_6$D$_6$, 150 MHz) δ (ppm): -4.6, -4.5, -4.4, -4.0, 14.4, 18.5, 19.0, 19.1, 20.6, 26.3, 26.6, 35.8, 62.3, 62.6, 72.4, 73.7, 84.0, 94.8, 100.7, 114.0, 114.8, 122.8, 128.4, 128.6, 128.8, 132.4, 136.3, 140.1, 145.7, 157.8, 158.6, 185.0, 188.1.

(*R*)-10-hydroxy-8-((2*S*, 4*R*, 5*S*)- 5-(*tert*-butyldimethylsiloxy)- 4-(*tert*-butyldimethylsiloxymethyl)-1,3-dioxan-2-yl)- 3-methyl-1-propyl- 1*H*-benzo[*g*]isochromene-6,9-dione (*ent*-1.53)

ent-1.51 → **ent-1.53**

1) Raney Ni, EtOH, rt, 10 min
2) O$_2$, salcomine, EtOH, rt, 1 h, 13 % (2 steps)

To a solution of **ent-1.51** 200.7 mg (0.239 mmol) in EtOH 20 ml was added Raney Ni (W4) 3.4 g, and the mixture was stirred for 10 minutes. The mixture was filtrated and washed with EtOH. Salcomine 32.0 mg (0.0984 mmol) was added to the mixture, which was vigorously stirred for an hour under oxygen atmosphere. The reaction mixture was evaporated in vacuo, and the residue was purified by subsequent seven column chromatography (*n*-hexane-1,2-dichloroethane 3：1) to give the naphthoquinone **ent-1.53** as red solids (19.4 mg, 0.0310 mmol, 13 %).

R_f: 0.40 (*n*-hexane-toluene 1：1)

HR-MS (FAB+): calcd. for C$_{34}$H$_{53}$O$_8$Si$_2$: 645.3279; found 645.3271.
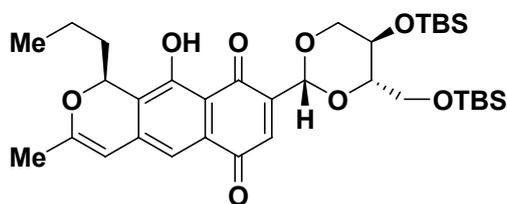
mp: 162.0-163.5 °C

IR (cm^{-1}, KBr): 2956, 2930, 2857, 1650, 1602, 1296, 1253, 1108, 837, 778.

^{1}H NMR (C$_6$D$_6$, 600 MHz) δ (ppm): -0.04 (3H, s, -Si-*Me$_2$*), 0.06 (3H, s, -Si-*Me$_2$*), 0.09 (3H, s, -Si-*Me$_2$*), 0.13 (3H, s, -Si-*Me$_2$*), 0.90 (3H, t, J = 7.0 Hz, H-13), 0.91 (9H, s, -Si-*t-Bu*), 1.00 (9H, s, -Si-*t-Bu*), 1.40-1.49 (1H, m, H-12), 1.51-1.59 (2H, m, H-11 & H-12), 1.60 (3H, br s, H-14), 1.95-2.02 (1H, m, H-11), 3.41-3.45 (1H, m, H-5'), 3.43 (1H, dd, J = 10.5 & 10.5 Hz, H-3'), 3.82 (1H, dd, J = 11.5 & 1.8 Hz, H-6'), 3.88 (1H, dd, J = 11.5 & 3.5 Hz, H-6'), 3.95 (1H, ddd, J = 10.5, 9.3 & 5.5 Hz, H-4'), 4.10 (1H, dd, J = 10.5 & 5.5 Hz, H-3'), 5.14 (1H, br s, H-4), 5.69 (1H, br s, H-1'), 5.78 (1H, dd, J = 9.5 & 3.3 Hz, H-1), 7.19 (1H, s, H-5), 7.37 (1H, br s, H-7), 12.6 (1H, d, J = 0.5 Hz, -OH).

^{13}C NMR (C$_6$D$_6$, 150 MHz) δ (ppm): -4.6, -4.5, -4.4, -4.0, 14.4, 18.5, 19.0, 19.1, 20.6, 26.3, 26.3, 26.6, 26.6, 35.8, 62.3, 62.7, 72.4, 73.7, 84.0, 94.8, 94.8, 100.7, 114.0, 114.8, 122.8, 128.4, 128.6, 128.8, 132.4, 136.3, 140.1, 145.7, 157.8, 158.6, 185.0, 188.1.

(*S*)-10-hydroxy-8-((2*R*, 4*S*, 5*R*)- 5-(*tert*-butyldimethylsiloxy)-
4-(*tert*-butyldimethylsiloxymethyl)-1,3-dioxan-2-yl)- 3-methyl-1-propyl-
1*H*-benzo[*g*]isochromene-6,9-dione (**1.53**)

1.53

*R*f: 0.40 (*n*-hexane-toluene 1 : 1)

HR-MS (FAB+): calcd. for $C_{34}H_{53}O_8Si_2$: 645.3279; found 645.3271.

mp: 162.0-163.5 °C

IR (cm^{-1}, KBr): 2956, 2930, 2857, 1650, 1602, 1296, 1253, 1108, 837, 778.

^{1}H NMR (C$_6$D$_6$, 600 MHz) δ (ppm): -0.04 (3H, s, -Si-*Me₂*), 0.06 (3H, s, -Si-*Me₂*), 0.09 (3H, s, -Si-*Me₂*), 0.13 (3H, s, -Si-*Me₂*), 0.90 (3H, t, *J* = 7.0 Hz, H-13), 0.91 (9H, s, -Si-*t-Bu*), 1.00 (9H, s, -Si-*t-Bu*), 1.40-1.50 (1H, m, H-12), 1.50-1.58 (2H, m, H-11 & H-12), 1.60 (3H, d, *J* = 1.0 Hz, H-14), 1.94-2.03 (1H, m, H-11), 3.41-3.45 (1H, m, H-5'), 3.43 (1H, dd, *J* = 10.5 & 10.5 Hz, H-3'), 3.82 (1H, dd, *J* = 11.5 & 1.8 Hz, H-6'), 3.88 (1H, dd, *J* = 11.5 & 3.5 Hz, H-6'), 3.95 (1H, ddd, *J* = 10.5, 9.3 & 5.5 Hz, H-4'), 4.10 (1H, dd, *J* = 10.5 & 5.5 Hz, H-3'), 5.14 (1H, d, *J* = 1.0 Hz, H-4), 5.69 (1H, br s, H-1'), 5.78 (1H, dd, *J* = 9.5 & 3.3 Hz, H-1), 7.19 (1H, s, H-5), 7.37 (1H, br s, H-7), 12.6 (1H, br s, -OH).

^{13}C NMR (C$_6$D$_6$, 150 MHz) δ (ppm): -4.6, -4.5, -4.4, -4.0, 14.4, 18.5, 19.0, 19.1, 20.6, 26.27, 26.30, 26.56, 26.58, 35.8, 62.3, 62.7, 72.4, 73.7, 84.0, 94.81, 94.83, 100.7, 114.0, 114.8, 122.8, 128.4, 128.6, 128.8, 132.4, 136.3, 140.1, 145.7, 157.8, 158.6, 185.0, 188.1.

(*S*)-10-hydroxy-8-((2*S*, 4*R*, 5*S*)- 5-(*tert*-butyldimethylsiloxy)-
4-(*tert*-butyldimethylsiloxymethyl)-1,3-dioxan-2-yl)- 3-methyl-1-propyl-
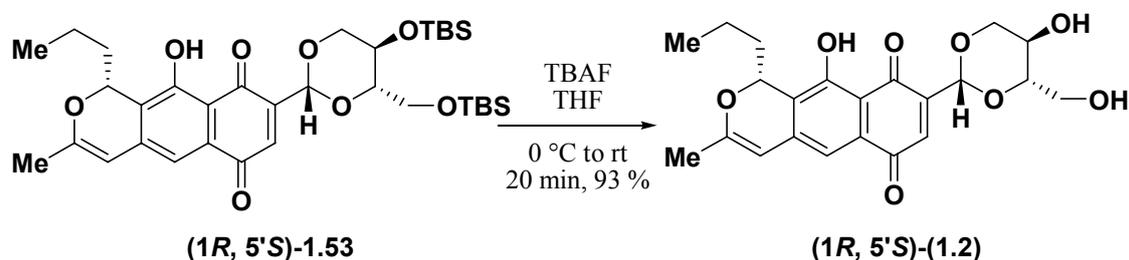1*H*-benzo[*g*]isochromene-6,9-dione ((**1***S*, **5'***R*)-**1.53**)
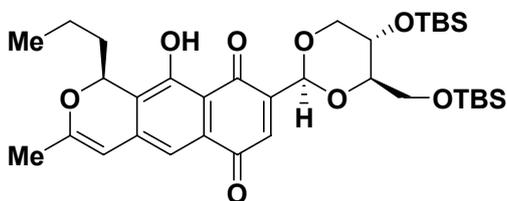
(**1***S*, **5'***R*)-**1.53**

*R*f: 0.40 (*n*-hexane-toluene 1 : 1)

^{1}H NMR (C$_6$D$_6$, 600 MHz) δ (ppm): -0.04 (3H, s, -Si-*Me₂*), 0.06 (3H, s, -Si-*Me₂*), 0.11 (3H, s, -Si-*Me₂*), 0.14 (3H, s, -Si-*Me₂*), 0.90 (3H, t, *J* = 7.0 Hz, H-13), 0.91 (9H, s, -Si-*t-Bu*), 1.00 (9H, s, -Si-*t-Bu*),

1.40-1.49 (1H, m, H-12), 1.50-1.59 (2H, m, H-11 & H-12), 1.59 (3H, br s, H-14), 1.94-2.03 (1H, m, H-11), 3.42 (1H, dd, J = 10.5 & 10.5 Hz, H-3'), 3.44 (1H, ddd, J = 9.5, 3.5 & 1.8 Hz, H-5'), 3.83 (1H, dd, J = 11.5 & 1.8 Hz, H-6'), 3.89 (1H, dd, J = 11.5 & 3.5 Hz, H-6'), 3.96 (1H, ddd, J = 10.5, 9.5 & 5.5 Hz, H-4'), 4.09 (1H, dd, J = 10.5 & 5.5 Hz, H-3'), 5.14 (1H, br s, H-4), 5.69 (1H, s, H-1'), 5.78 (1H, dd, J = 9.5 & 3.5 Hz, H-1), 7.19 (1H, s, H-5), 7.37 (1H, s, H-7), 12.60 (1H, s, -OH).

^{13}C NMR (C$_6$D$_6$, 150 MHz) δ (ppm): -4.6, -4.5, -4.4, -4.0, 14.4, 18.5, 19.0, 19.1, 20.6, 26.3, 26.6, 35.8, 62.3, 62.6, 72.4, 73.7, 84.0, 94.8, 100.7, 114.0, 114.8, 122.8, 128.4, 128.6, 128.8, 132.4, 136.3, 140.1, 145.7, 157.8, 158.6, 185.0, 188.1.

(R)-10-hydroxy-8-((2R, 4S, 5R)- 5-hydroxy-4-(hydroxymethyl)-1,3-dioxan-2-yl)- 3-methyl-1-propyl-1H-benzo[g]isochromene-6,9-dione ((1R,5'S)-(1.2))

(1R, 5'S)-1.53 → **(1R, 5'S)-(1.2)**

To a solution of the naphthoquinone **(1R, 5'S)-1.53** 11.8 mg (0.0183 mmol) in THF 1.3 ml was added TBAF 1.0 M solution in THF 55 µl at 0 ° C. After 20 minutes, the reaction mixture was quenched with tenfold dilute acetic acid 31 µl with THF. The mixture was evaporated in vacuo, and the residue was purified by silica gel column chromatography (n-hexane-2-propanol 4 : 1 including 1% AcOH) to give **(1R, 5'S)-(1.2)** as red solids (7.1 mg, 0.0170 mmol, 93 %).

R_f: 0.36 (n-hexane-2-propanol 7 : 3)

HR-MS (EI+): calcd. for C$_{22}$H$_{24}$O$_8$: 416.1471; found 416.1471.

mp : 164.5-165.8 ° C (decomp.)

IR (cm^{-1}, KBr): 3400, 2958, 2872, 1641, 1599, 1386, 1296, 1089, 1033.

^{1}H NMR (CDCl3, 600 MHz) δ (ppm): 0.96 (3H, t, J = 7.0 Hz, H-13), 1.43-1.60 (3H, m, H-11 & H-12), 1.96 (3H, d, J = 0.5 Hz, H-14), 1.91 (1H, t, J = 6.0 Hz, -OH), 1.95-2.02 (1H, m, H-11), 2.22 (1H, d, J = 5.0 Hz, -OH), 3.63 (1H, dd, J = 10.5 & 10.5 Hz, H-3'), 3.74 (1H, ddd, J = 9.3, 4.5 & 4.5 Hz, H-5'), 3.89-3.97 (3H, m, H-4' & H-6'), 4.31 (1H, dd, J = 10.8 & 5.5 Hz, H-3'), 5.60 (1H, d, J = 0.5 Hz, H-4), 5.64 (1H, dd, J = 8.5 & 3.5 Hz, H-1), 5.72 (1H, d, J = 0.8 Hz, H-1'), 7.07 (1H, d, J = 0.8 Hz, H-7), 7.13 (1H, s, H-5), 12.17 (1H, s, -OH).

^{13}C NMR (CDCl3, 150 MHz) δ (ppm): 13.8, 18.1, 20.4, 34.8, 62.6, 62.7, 70.9, 73.0, 81.4, 94.0, 100.0, 112.9, 114.4, 122.4, 131.3, 135.6, 139.7, 144.3, 157.1, 158.6, 184.7, 186.9.

(*R*)-10-hydroxy-8-((2*S*, 4*R*, 5*S*)- 5-hydroxy-4-(hydroxymethyl)-
1,3-dioxan-2-yl)- 3-methyl-1-propyl-1*H*-benzo[*g*]isochromene-6,9-dione ((1*R*,5'*R*)-(1.2))

ent-1.53   **(1*R*, 5'*R*)-(1.2)**
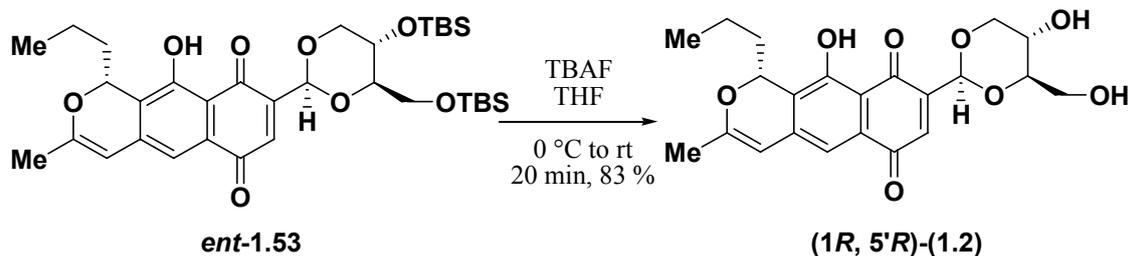
To a solution of the naphthoquinone **ent-1.53** 11.6 mg (0.0180 mmol) in THF 1.2 ml was added TBAF 1.0 M solution in THF 55 µl at 0 ° C. After 20 minutes, the reaction mixture was quenched with tenfold dilute acetic acid 31 µl with THF. The mixture was evaporated in vacuo, and the residue was purified by silica gel column chromatography (n-hexane-2-propanol 4 : 1 including 1% AcOH) to give **(1*R*, 5'*R*)-(1.2)** as red solids (6.2 mg, 0.0149 mmol, 83 %).

R_f: 0.36 (*n*-hexane-2-propanol 7 : 3)

HR-MS (EI+): calcd. for $C_{22}H_{24}O_8$: 416.1471; found 416.1466.

mp : 138.0-139.5 ° C (decomp.)

IR (cm^{-1}, KBr): 3380, 2959, 2873, 1644, 1601, 1387, 1296, 1089, 1032, 889.

^{1}H NMR (CDCl3, 600 MHz) δ (ppm): 0.96 (3H, t, J = 7.0 Hz, H-13), 1.43-1.63 (3H, m, H-11 & H-12), 1.96 (3H, d, J = 0.6 Hz, H-14), 1.94-2.03 (2H, m, H-11 & -OH), 2.32-2.36 (1H, m, -OH), 3.65 (1H, dd, J = 10.5 & 10.5 Hz, H-3'), 3.74 (1H, ddd, J = 9.0, 4.5 & 4.5 Hz, H-5'), 3.89-3.96 (3H, m, H-4' & H-6'), 4.31 (1H, dd, J = 10.5 & 5.5 Hz, H-3'), 5.60 (1H, d, J = 0.6 Hz, H-4), 5.65 (1H, dd, J = 9.5 & 3.5 Hz, H-1), 5.72 (1H, d, J = 1.0 Hz, H-1'), 7.07 (1H, d, J = 1.0 Hz, H-7), 7.13 (1H, s, H-5), 12.17 (1H, s, -OH).

^{13}C NMR (CDCl3, 150 MHz) δ (ppm): 13.8, 18.1, 20.4, 34.9, 62.7, 62.7, 70.9, 73.0, 81.4, 94.0, 100.0, 112.9, 114.4, 122.4, 131.3, 135.6, 139.7, 144.3, 157.1, 158.6, 184.7, 186.9.

(*S*)-10-hydroxy-8-((2*R*, 4*S*, 5*R*)- 5-hydroxy-4-(hydroxymethyl)-
1,3-dioxan-2-yl)- 3-methyl-1-propyl-1*H*-benzo[*g*]isochromene-6,9-dione ((1*S*,5'*S*)-(1.2))

(1*S*,5'*S*)-(1.2)

R_f: 0.36 (*n*-hexane-2-propanol 7 : 3)

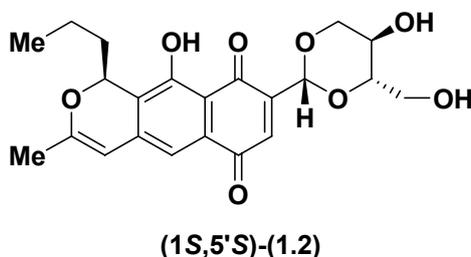HR-MS (EI+): calcd. for $C_{22}H_{24}O_8$: 416.1471; found 416.1466.

mp : 138.0-139.5 ° C (decomp.)

IR (cm⁻¹, KBr): 3380, 2959, 2873, 1644, 1601, 1387, 1296, 1089, 1032, 889.

¹H NMR (CDCl3, 600 MHz) δ (ppm): 0.96 (3H, t, J = 7.0 Hz, H-13), 1.43-1.62 (3H, m, H-11 & H-12), 1.96 (3H, d, J = 0.6 Hz, H-14), 1.93-2.03 (2H, m, H-11 & -OH), 2.28 (1H, d, J = 4.8 Hz, -OH), 3.65 (1H, dd, J = 10.5 & 10.5 Hz, H-3'), 3.74 (1H, ddd, J = 9.0, 4.5 & 4.5 Hz, H-5'), 3.89-3.96 (3H, m, H-4' & H-6'), 4.31 (1H, dd, J = 10.5 & 5.5 Hz, H-3'), 5.60 (1H, d, J = 0.6 Hz, H-4), 5.65 (1H, dd, J = 9.5 & 3.5 Hz, H-1), 5.72 (1H, d, J = 1.0 Hz, H-1'), 7.07 (1H, d, J = 1.0 Hz, H-7), 7.12 (1H, s, H-5), 12.17 (1H, s, -OH).

¹³C NMR (CDCl3, 150 MHz) δ (ppm): 13.8, 18.1, 20.4, 34.9, 62.6, 62.7, 70.9, 73.0, 81.5, 94.0, 100.0, 112.9, 114.4, 122.4, 131.3, 135.6, 139.7, 144.3, 157.1, 158.6, 184.7, 186.9.

(*S*)-10-hydroxy-8-((2*S*, 4*R*, 5*S*)- 5-hydroxy-4-(hydroxymethyl)-1,3-dioxan-2-yl)- 3-methyl-1-propyl-1*H*-benzo[*g*]isochromene-6,9-dione ((1*S*,5'*R*)-(1.2))
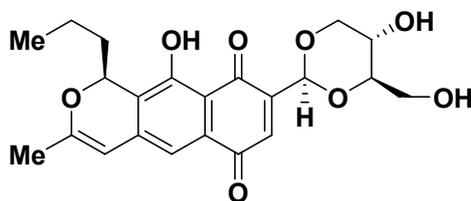

(1S,5'R)-(1.2)

R_f: 0.36 (*n*-hexane-2-propanol 7 : 3)

¹H NMR (CDCl3, 600 MHz) δ (ppm): 0.96 (3H, t, J = 7.0 Hz, H-13), 1.43-1.60 (3H, m, H-11 & H-12), 1.96 (3H, d, J = 0.5 Hz, H-14), 1.91 (1H, t, J = 6.0 Hz, -OH), 1.95-2.02 (1H, m, H-11), 2.22 (1H, d, J = 5.0 Hz, -OH), 3.63 (1H, t, J = 10.5 Hz, H-3'), 3.74 (1H, dt, J = 9.3 & 4.5 Hz, H-5'), 3.89-3.97 (3H, m, H-4' & H-6'), 4.31 (1H, dd, J = 10.8 & 5.5 Hz, H-3'), 5.60 (1H, d, J = 0.5 Hz, H-4), 5.64 (1H, dd, J = 8.5 & 3.5 Hz, H-1), 5.72 (1H, d, J = 0.8 Hz, H-1'), 7.07 (1H, d, J = 0.8 Hz, H-7), 7.13 (1H, s, H-5), 12.17 (1H, s, -OH).

¹³C NMR (CDCl3, 150 MHz) δ (ppm): 13.8, 18.1, 20.4, 34.8, 62.6, 62.7, 70.9, 73.0, 81.4, 94.0, 100.0, 112.9, 114.4, 122.4, 131.3, 135.6, 139.7, 144.3, 157.1, 158.6, 184.7, 186.9.

Natural K1115 B₁s (1.2)
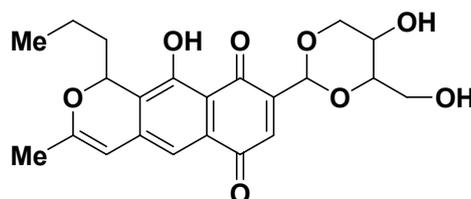
K1115 B₁s

¹H NMR (CDCl₃, 600 MHz) δ (ppm): 0.957 [0.959] (3H, t, J = 7.0 Hz, H-13), 1.43-1.61 (3H, m, H-11 & H-12), 1.87-1.95 (1H, m, -OH), 1.96 (3H, s, H-14), 1.95-2.03 (1H, m, H-11), 2.21-2.26 (1H, m, -OH),

3.65 (1H, dd, J = 10.5 & 10.5 Hz, H-3'), 3.74 (1H, ddd, J = 9.3, 4.5 & 4.5 Hz, H-5'), 3.89-3.97 (3H, m, H-4' & H-6'), 4.306 (1H, dd, J = 10.5 & 5.5 Hz, H-3'), [4.311], 5.60 (1H, s, H-4), 5.64 [5.65] (1H, dd, J = 9.5 & 3.5 Hz, H-1), 5.72 (1H, s, H-1'), 7.07 (1H, s, H-7), 7.13 (1H, s, H-5), 12.171 [12.174] (1H, s, -OH). ^{13}C NMR (CDCl3, 150 MHz) δ (ppm): 13.8, 18.1, 20.4, 34.8, 62.6, 62.7, 70.9, 73.0, 81.4, 94.0, 100.0, 112.9, 114.4, 122.4, 131.3, 135.6, 139.7, 144.3, 157.1, 158.6, 184.7, 186.9.

* [ ]: K1115B$_{1\beta}$

Section 4. The First Total Synthesis and the Structural Determination of TMC-66.

1,4-Dihydro-5-methoxy-1,4-dioxonaphthalen-2-yl acetate (2.32)
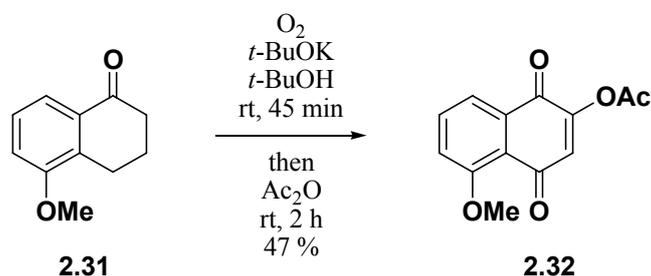
A solution of compound **2.31** 10.1 mg (57.3 µmol) in t-BuOH 500 µl was stirred for 15 minutes, then potassium *tert*-butoxide 32.2 mg (287 µmol) was added and the mixture was stirred at room temperature. After stirring for 45 minutes, acetic anhydride 500 µl was added and the resulting mixture was stirred at room temperature for 2 hours. After evaporation in vacuo, the crude residue was purified by flash column chromatography (hexane-EtOAc 2 : 1) to yield the naphthoquinone **2.32** as a yellow solid (6.6 mg, 26.8 µmol, 47 %)

R_f: 0.38 (hexane-EtOAc 1 : 1).

HR-MS (FAB+): calcd. for $C_{13}H_{11}O_5$: 247.0607, found 247.0605 [M+H]+.
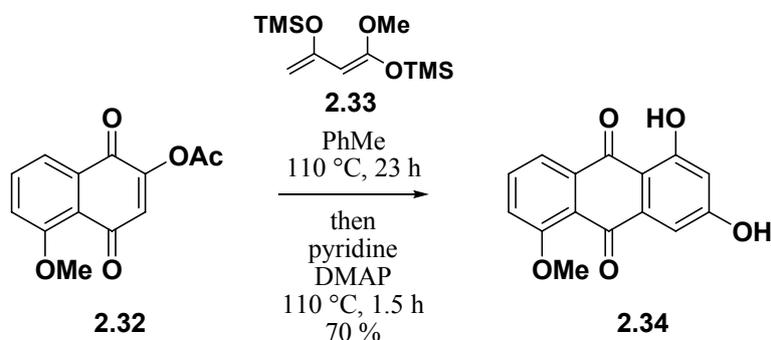
mp 136.5-140.5 °C (decomp.)

IR (cm^{-1}, KBr): 1770, 1658, 1587, 1184.

1H NMR (CDCl$_3$, 600MHz at 25 °C) δ (ppm): 2.37 (3H, s, Me of Ac), 4.01 (3H, s, OMe), 6.66 (1H, s, H-7a), 7.34 (1H, dd, J = 8.5 & 1.0 Hz, H-10), 7.69 (1H, dd, J = 8.5 & 7.5 Hz, H-11), 7.78 (1H, dd, J = 7.5 & 1.0 Hz, H-12).

13C NMR (CDCl$_3$, 150 Hz at 25 °C) δ (ppm): 20.5, 56.6, 118.6, 119.5, 119.8, 128.0, 133.2, 135.0, 152.3, 159.7, 167.7, 178.9, 183.7

1,3-Dihydroxy-5-methoxyanthracene-9,10-dione (2.34)

Diene **2.33** 93.0 ml (357 µmol) was added to a solution of naphtoquinone **2.32** 29.3 mg (119 µmol) in toluene, and the mixture was stirred at 110 °C for 23 hours. Pyridine 11.5 ml (143 µmol) and *N,*

N-dimethylaminopyridine 6.7 mg (59.5 µmol) was added to the reaction mixture, which was stirred for 1.5 hours at the same temperature. After evaporation in vacuo, acetone 400 µl was added to the residue. The resulting precipitates were collected by filtration and dried in vacuo to yield the compound **2.34** as a yellow solid (22.5 mg, 83.3 µmol, 70%).

R_f : 0.34 (toluene-EtOAc 2：1)
HR-MS (FAB+): calcd. for $C_{15}H_{11}O_5$：271.0607, found 271.0598 [M+H]$^+$.
mp：300 °C<
IR (cm^{-1}, KBr): 1627, 1340, 1288, 1160.
^{1}H NMR (DMSO-d_6, 600 MHz at 25 °C) δ (ppm): 3.91 (3H, s, OMe), 6.50 (1H, d, J = 2.5 Hz, H-14a), 7.00 (1H, d, J = 2.5 Hz, H-7), 7.53 (1H, d, J = 8.5 Hz & 1.0 Hz, H-10), 7.77 (1H, dd, J = 7.5 Hz & 1.0 Hz, H-12), 7.80 (1H, dd, J = 8.5 Hz & 7.5 Hz, H-11), 11.23 (1H, s, HO-6a), 12.62 (1H, s, HO-14).
^{13}C NMR (DMSO-d_6, 150 MHz at 25 °C) δ (ppm): 56.86, 107.08, 108.38, 109.35, 119.06, 119.57, 120.83, 135.40, 136.09, 137.07, 160.62, 164.75, 166.02, 180.78, 186.24

9,10-Dihydro-1-hydroxy-5-methoxy-9,10-dioxoanthracen-3-yl trifluoromethanesulfonate (2.35)
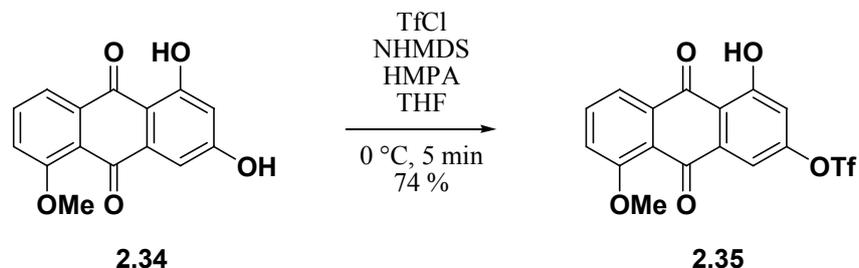
To a solution of the compound **2.34** 1.02g (3.77 mmol) in THF 20 ml and HMPA 2 ml was added NHMDS 2.19 ml (1.90 M in THF, 4.15 mmol) at 0 °C, and the mixture was stirred at the same temperature. After stirring for 15 minutes, trifluoromethanesulfonyl chloride 456 µl (4.34 mmol) was added and stirred for 45 minutes at　0 °C. The reaction mixture was quenched with 1M HCl 5 ml and the mixture was evaporated in vacuo. Then, H$_2$O 10 ml was added, and the mixture was extracted with ethyl acetate (2×30 ml). The combined extracts were evaporated in vacuo. The crude residue was purified by flash column chlomatography (toluene-ethyl acetate 20:1) to yield the monotriflate **2.35** as a yellow solid (1.11g, 2.77 mmol, 74%).

R_f : 0.45 (toluene-EtOAc 5：1)
HR-MS (FAB+): calcd. for $C_{16}H_{10}F_3O_7S$：403.0099, found 403.0076 [M+H]$^+$.
mp：154.0-155.0 °C
IR (cm^{-1}, KBr): 3085, 1672, 1645, 1241, 1209.
^{1}H NMR (CDCl$_3$, 600 MHz at 25 °C) δ (ppm): 4.07 (3H, s, OMe), 7.15 (1H, d, J = 2.5 Hz, H-14a), 7.41 (1H, d, J = 8.5 Hz & 1.0 Hz, H-10), 7.65 (1H, d, J = 2.5 Hz, H-7), 7.79 (1H, dd, J = 8.5 Hz & 7.5 Hz, H-11), 7.99 (1H, dd, J = 7.5 Hz & 1.0 Hz, H-12), 12.60 (1H, s, HO-14).
^{13}C NMR (CDCl$_3$, 150 MHz at 25 °C) δ (ppm): 56.7, 112.1, 115.2, 115.5, 118.6 (q, J = 321 Hz, Tf), 119.0, 119.8, 120.8, 134.8, 135.8, 137.2, 154.5, 160.9, 163.6, 179.6, 187.7

5,7-Bis(trifluoromethanesulfoxy)-2,2-dimethyl-4*H*-benzo[*d*][1,3]dioxin-4-one (2.52)
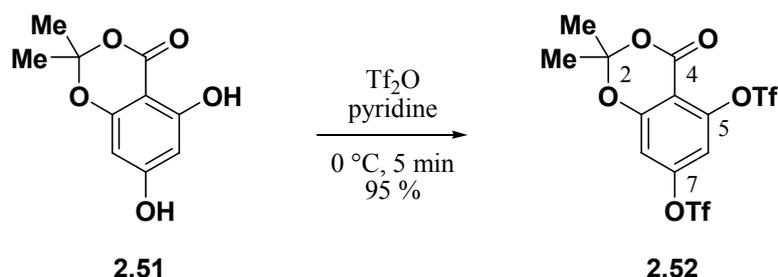
To a solution of the compound **2.51** 40.7 mg (194 µmol) in pyridine 400 µl was added trifluoromethanesulfonic anhydride 81.5 µl, (484 µmol) and the mixture was stirred for 5 minutes. The reaction mixture was quenched with sat. NaHCO₃ 200 µl and extracted with ethyl acetate (3×600µl). The combined extracts was evaporated in vacuo. The crude residue was purified by flash column chromatography (hexane-ethyl acetate 4:1) to yield the bistriflate **2.52** as a colorless solid (87.2 mg, 184 µmol, 95%).

R_f: 0.74 (hexane-EtOAc 2 : 1)
HR-MS (FAB+): calcd. for $C_{12}H_9F_6O_9S_2$: 474.9592, found 474.9572 [M+H]$^+$.
mp : 95.10-95.6 °C
IR (cm^{-1}, KBr): 1752, 1621, 1429, 1211, 1135.
^{1}H NMR (CDCl₃, 600 MHz at 25 °C) δ (ppm): 1.80 (6H, s, C$\underline{Me}_2$), 6.93 (1H, d, J = 2.5 Hz, H-4), 7.04 (1H, d, J = 2.5 Hz, H-14b).
^{13}C NMR (CDCl₃, 150 MHz at 25 °C) δ (ppm): 25.5, 107.8, 108.2, 110.7, 111.2, 118.5 (q, J = 321 Hz, Tf), 118.7 (q, J = 321 Hz, Tf), 149.6, 153.2, 155.8, 158.3

2,2-Dimethyl-7-(2-(trimethylsilyl)ethynyl)-4-oxo-4*H*-benzo[*d*][1,3]dioxin-5-yl trifluoromethanesulfonate (2.53)
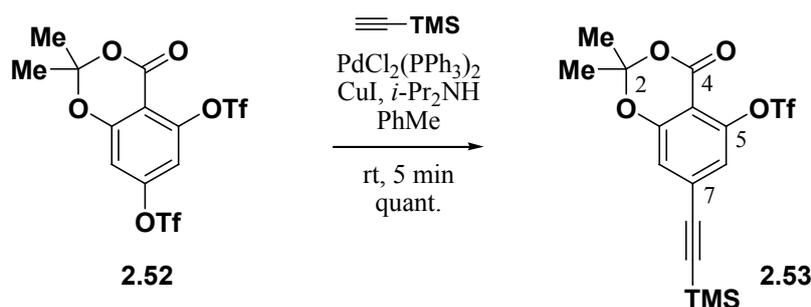
To a solution of compound **2.52** 59.9 mg (126 µmol) in toluene 1.2 ml was added diisopropylamine 25.0µl (253 µmol), PdCl₂(PPh₃)₂ 8.9 mg (12.6 µmol), and CuI 2.4 mg (12.6 µmol). The mixture was stirred at room temperature for 5 minutes, and trimethylsilylacetylene 19.6µl (139 µmol) was added. After stirring for more 5 minutes, the reaction mixture was evaporated in vacuo. The crude residue was purified by flash column chromatography (hexane-ethyl acetate 3:1) to yield the compound **2.53** as a colorless solid (53.4 mg, 126 µmol, quant.).

R_f: 0.60 (hexane-EtOAc 3 : 1)

HR-MS (FAB+): calcd. for $C_{16}H_{18}F_3O_6SSi$: 423.0545, found 423.0527 [M+H]⁺.

mp : 94.4-96.0 °C

IR (cm⁻¹, KBr): 1751, 1621, 1558, 1207, 1139.

¹H NMR (CDCl₃, 600 MHz at 25 °C) δ (ppm): 0.27 (9H, s, SiMe₃), 1.75 (6H, s, CMe₂), 7.02 (1H, d, J = 1.5 Hz, H-4), 7.10 (1H, d, J = 1.5 Hz, H-14b).

¹³C NMR (CDCl₃, 150 MHz at 25 °C) δ (ppm): -0.5, 25.5, 101.1, 102.4, 107.0, 107.9, 118.7 (q, J = 322 Hz, Tf), 119.6, 120.8, 131.5, 148.4, 156.7, 157.1.

2,2-Dimethyl-7-(2-(trimethylsilyl)ethynyl)-5-(2-oxopropyl)-4H-benzo[d][1,3]dioxin-4-one (2.54)
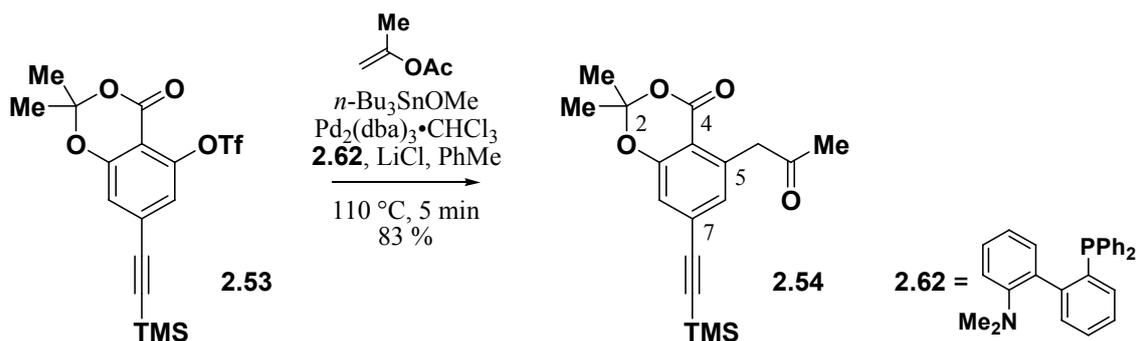
To a solution of isopropenyl acetate 1.07 ml (9.72 µmol) in toluene 1.2 ml was added tributyltin methoxide 1.96 ml (6.81 µmol), and the mixture was heated at 110 °C for 20 minutes. After cooling to ambient temperature, Pd₂(dba)₃ · CHCl₃ 152 mg (166 µmol), 2-diphenylphosphino-2'-(N,N-dimethylamino)biphenyl (2.62) 381 mg (999 µmol), and LiCl 282 mg (6.66 µmol) were added to the mixture. After heating at 110 °C for 5 minutes, a solution of compound 2.53 703 mg (1.66 µmol) in toluene 2.4 ml was added. After 5 minutes at the elevated temperature, the reaction mixture was evaporated in vacuo. The crude residue was purified by flash column chromatography (hexane-ethyl acetate 7:1) to yield the compound 2.54 as a colorless solid (456 mg, 1.38 µmol, 83%).

R_f: 0.45 (hexane-EtOAc 2 : 1)

HR-MS (FAB+): calcd. for $C_{18}H_{23}O_4Si$: 331.1366, found 331.1357 [M+H]⁺.

mp : 102.0-103.3 °C

IR (cm⁻¹, KBr): 2962, 2161, 1733, 1612, 1284, 844.

¹H NMR (CDCl₃, 600 MHz at 25 °C) δ (ppm): 0.25 (9H, s, SiMe₃), 1.71 (6H, s, CMe₂), 2.30 (3H, s, Me-21), 4.14 (2H, s, H-19), 6.92 (1H, d, J = 1.5 Hz, H-4), 6.99 (1H, d, J = 1.5 Hz, H-14b).

¹³C NMR (CDCl₃, 150 MHz at 25 °C) δ (ppm): -0.3, 25.6, 30.2, 48.8, 99.3, 102.9, 105.9, 112.6, 119.6, 129.6, 130.1, 138.8, 156.7, 160.6, 204.7.

(3*R*,10a*R*)-benzyl 8-Ethynyl-3,5,10,10a-tetrahydro-6-hydoxy-10a-methyl-5-oxo-2*H*-oxazolo[3,2-*b*]isoquinoline-3-carboxylate (**2.48**)
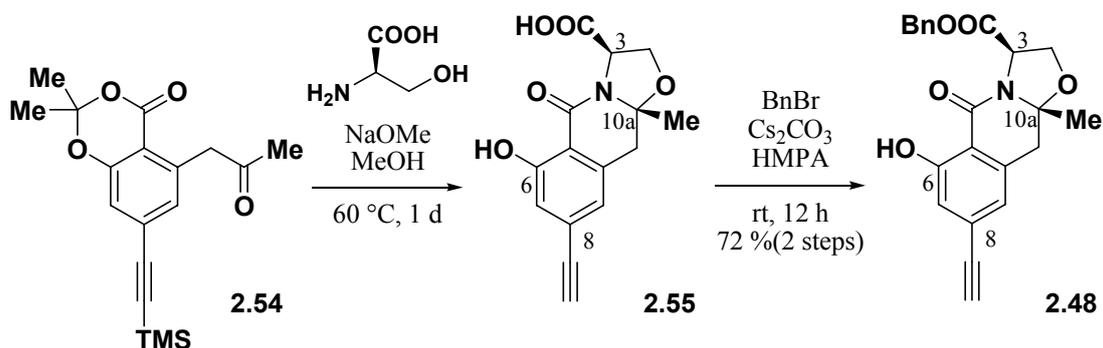
To a solution of D-serine 2.0 mg (19 µmol) in MeOH 100 µl was added sodium methoxide 5.2 mg (16 µmol), and the reaction mixture was stirred at 0 °C for 10 minutes. Compound **2.54** 5.2 mg (16 µmol) was added, and the reaction mixture was heated at 60 °C. After stirring for 1 day, the reaction mixture was evaporated in vacuo to give the crude tricyclic **2.55** as a single diastereomer, which was used in the next step without further purification.

To a solution of the crude tricyclic **2.55** in HMPA 100 µl were added Cs_2CO_3 10 mg (32 µmol) and BnBr 3.8 µl (32 µmol), and the mixture was stirred at room temperature for 12 hours. The reaction mixture was quenched with H_2O 100 µl and extracted with toluene (3×200 µl). The combined extracts were evaporated in vacuo. The crude residue was purified by flash column chromatography (hexane-ethyl acetate 4:1) to yield the compound **2.48** as a colorless solid (4.4 mg, 12 µmol, 72%).

R_f : 0.50 (hexane-EtOAc 2 : 1)
HR-MS (FAB+): calcd. for $C_{22}H_{20}NO_5$: 378.1341, found 378.1361 [M+H]+.
$[\alpha]_D^{28}$ -48.7 ° (c, 1.02, CHCl$_3$)
mp : 107.9-108.6 °C
IR (cm^{-1}, KBr): 3282, 2958, 2111, 1749, 1641, 1465, 1189.
^{1}H NMR (CDCl$_3$, 600 MHz at 25 °C) δ (ppm): 1.46 (3H, d, J = 1.0 Hz, Me-21) , 3.05 (1H, d, J = 15.0 Hz, H-19), 3.11 (1H, dq, J = 15.0 & 1.0 Hz, H-19), 3.18 (1H, s, H-6), 4.16 (1H, dd, J = 9.0 & 7.0 Hz, H-17), 4.50 (1H, dd, J = 9.0 & 8.5 Hz, H-17), 4.83 (1H, dd, J = 8.5 & 7.0 Hz, H-16), 5.25 (1H, d, J = 12.5 Hz, O-C$\underline{H}_2$-Ph), 5.27 (1H, d, J = 12.5 Hz, O-C$\underline{H}_2$-Ph), 6.82 (1H, br s, H-4), 6.98 (1H, br s, H-14b), 7.32-7.43 (5H, m, Ph), 11.51 (1H, s, HO-1).
^{13}C NMR (CDCl$_3$, 150 MHz at 25 °C) δ (ppm): 22.6, 40.6, 56.4, 67.5, 67.7, 79.8, 82.4, 94.3, 110.6, 120.1, 122.4, 128.1, 128.3, 128.6, 128.7, 134.9, 135.9, 160.8, 164.3, 169.3.

6-Ethynyl-8-hydroxy-3-methyl-1*H*-isochromen-1-one (2.56)
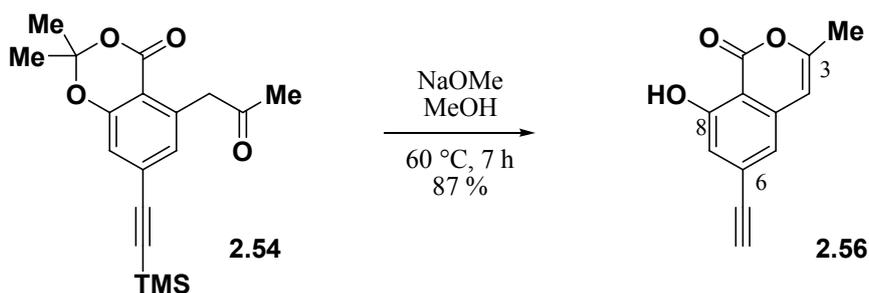
To a solution of compound **2.54** 29.6 mg (89.6 µmol) in methanol 890 µl was added sodium methoxide 4.8 mg (89.6 µmol), and the mixture was stirred at 60 °C. After stirring for 7 hours, the reaction mixture was evaporated in vacuo. Then, H_2O 250 µl and sat. $KHSO_4$ 20 µl were added , and the mixture was extracted with ethyl acetate (500 µl, then 300 µl). The combined extracts were evaporated in vacuo. The crude residue was purified by flash column chromatography (hexane-ethyl acetate 3:1) to yield the compound **2.56** as a colorless solid (15.5 mg, 77.4 µmol, 86%).

R_f : 0.45 (hexane-EtOAc 2 : 1)
FAB-MS : 201 [M+H]$^+$.
^{1}H NMR (CDCl$_3$, 600 MHz at 25 °C) δ (ppm): .2.28 (3H, br s, Me), 3.26 (1H, s, H-6), 6.21 (1H, br s, H-19), 6.90 (1H, br s, H-14b), 7.00 (1H, br s, H-4), 10.95 (1H, s, HO-1), H-6, H-14b, and H-19 are interchangeable.
^{13}C NMR (CDCl$_3$, 150 MHz at 25 °C) δ (ppm): 19.4, 80.9, 82.2, 104.2, 105.7, 117.8, 118.7, 130.9, 137.9, 154.7, 161.3, 166.3.

(3*R*,10a*R*)-Benzyl 3,5,10,10a-tetrahydro-8-(2-(9,10-dihydro-1-hydroxy-5-methoxy-9,10-dioxoanthracen-3-yl)ethynyl)-6-hydroxy-10a-methyl-5-oxo-2*H*-oxazolo[3,2-*b*]isoquinoline-3-carboxylate (2.59)
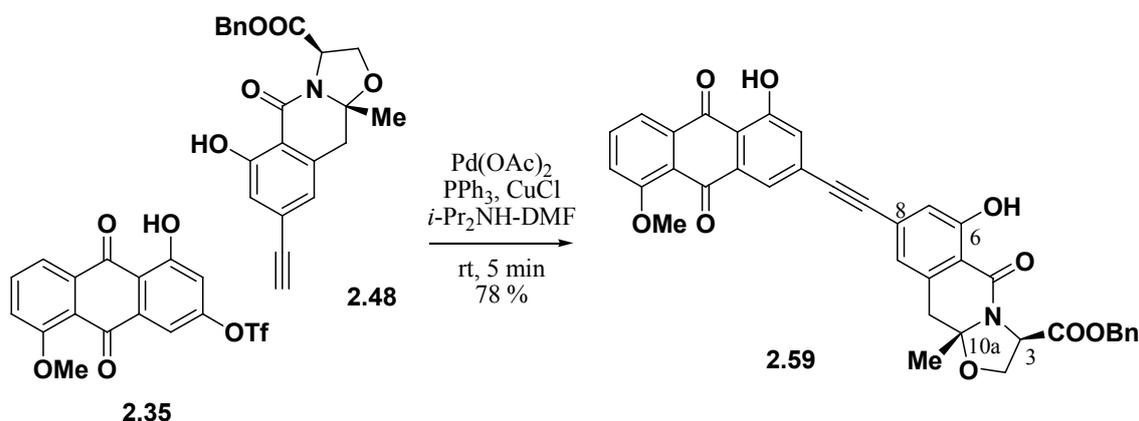
To a solution of compound **2.35** 20.5 mg (50.9 µmol) in DMF 400 µl and diisopropylamine 100 µl were added Pd(OAc)$_2$ 2.4 mg (10.2 µmol), PPh$_3$ 8.0 mg (30.5 µmol), and CuCl 5.0 mg (50.9 µmol). The mixture was stirred at room temperature for 5 minutes, and a solution of compound **2.48** 19.2 mg (50.9 µmol) in DMF 400 µl was added. After stirring for 5 minutes, the reaction mixture was

quenched with sat. KHSO$_4$ 100 µl and extracted with toluene (3×400 µl). The combined extracts were evaporated in vacuo. The crude residue was purified by flash column chromatography (toluene-ethyl acetate 7:1) to yield the hexacycle **2.59** as a yellow solid (25.0 mg, 39.7 µmol, 78%).

R_f : 0.42 (toluene-ethyl acetate 3:1)
HR-MS (FAB+): calcd. for C$_{37}$H$_{28}$NO$_9$: 630.1764, found 630.1768 [M+H]$^+$.
[α]$_D^{26}$ -43.2 ° (c, 0.99, CHCl$_3$)
mp : 230.3-237.4 °C
IR (cm^{-1}, KBr): 2923, 2210, 1749, 1670, 1635, 1284, 709.
^{1}H NMR (CDCl$_3$, 600 MHz at 25 °C) δ (ppm): 1.49 (3H, s, Me-21), 3.11 (1H, d, J = 15.0 Hz, H-19), 3.15 (1H, d, J = 15.0 Hz, H-19), 4.07 (3H, s, OMe), 4.18 (1H, dd, J = 9.0 & 7.0 Hz, H-17), 4.51 (1H, dd, J = 9.0 & 8.5 Hz, H-17), 4.85 (1H, dd, J = 8.5 & 7.0 Hz, H-16), 5.26 (1H, d, J = 13.0 Hz, O-C$\underline{H}_2$-Ph), 5.28 (1H, d, J = 13.0 Hz, O-C$\underline{H}_2$-Ph), 6.92 (1H, s, H-4), 7.06 (1H, s, H-14b), 7.35 (1H, d, J = 1.5 Hz, H-14a), 7.33-7.43 (6H, m, H-10 & Ph), 7.76 (1H, dd, J = 8.0 & 8.0 Hz, H-11), 7.87 (1H, d, J = 1.5 Hz, H-7), 7.98 (1H, dd, J = 8.0 & 1.0 Hz, H-12), 11.60 (1H, s, HO-1), 12.46 (1H, s, HO-14).
^{13}C NMR (CDCl$_3$, 150 MHz at 25 °C) δ (ppm): 22.6, 40.6, 56.5, 56.7, 67.5, 67.7, 90.3, 92.9, 94.3, 110.9, 115.4, 118.7, 119.6, 119.8, 121.3, 122.2, 122.5, 125.1, 128.1, 128.3, 128.6, 128.7, 131.2, 135.0, 135.3, 135.3, 136.1, 160.7, 160.9, 161.8, 164.3, 169.3, 181.0, 188.0.

(3R,10aR)-Benzyl 3,5,10,10a-tetrahydro-8-(2-(9,10-dihydro-1-hydroxy-5-methoxy-9,10-dioxoanthracen-3-yl)ethyl)-6-hydroxy-10a-methyl-5-oxo-2H-oxazolo[3,2-b]isoquinoline-3-carboxylate (**2.60**)
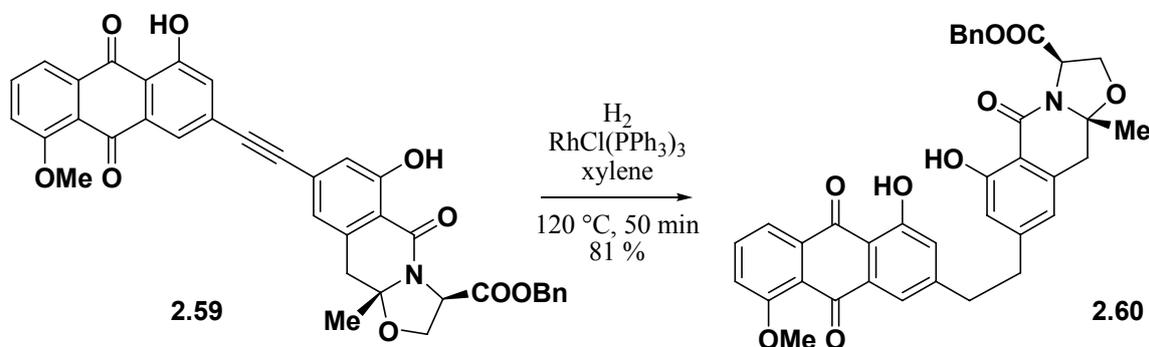
To a solution of the compound **2.59** 101 mg (160 µmol) in xylene 2.0 ml was added RhCl(PPh$_3$)$_3$ 57.0 mg (61.6 µmol), and the mixture was heated at 120 °C for 50 minutes. After cooling to ambient temperature, the reaction mixture was directly purified by flash column chromatography (toluene-ethyl acetate 7:1) to yield the compound **2.60** as a yellow solid (82.5 mg, 130 µmol, 81%).

R_f : 0.37 (toluene-ethyl acetate 3:1)
HR-MS (FAB+): calcd. for C$_{37}$H$_{32}$NO$_9$: 634.2077, found 634.2088 [M+H]$^+$.
[α]$_D^{27}$ -32.8 ° (c, 1.02, CHCl$_3$)
mp : 200.8-203.6 °C
IR (cm^{-1}, KBr): 2923, 1749, 1670, 1635, 1278, 796.
^{1}H NMR (CDCl$_3$, 600 MHz at 25 °C) δ (ppm): 1.46 (3H, s, Me-21), 2.90-2.95 (2H, m, H-5 & H-6), 2.98-3.05 (3H, m, H-5, H-6, & H-19), 3.10 (1H, d, J = 15.0 Hz, H-19), 4.05 (3H, s, OMe), 4.16 (1H, dd, J = 9.0 & 7.0 Hz, H-17), 4.48 (1H, dd, J = 9.0 & 8.5 Hz, H-17), 4.82 (1H, dd, J = 8.5 & 7.0 Hz, H-16),

5.25 (1H, d, J = 12.5 Hz, O-C$\underline{H}_2$-Ph), 5.27 (1H, d, J = 12.5 Hz, O-C$\underline{H}_2$-Ph), 6.51 (1H, s, H-4), 6.70 (1H, s, H-14b), 7.02 (1H, d, J = 1.5 Hz, H-14a), 7.32-7.42 (6H, m, H-10 & Ph), 7.62 (1H, d, J = 1.5 Hz, H-7), 7.74 (1H, dd, J = 8.0 & 7.5 Hz, H-11), 7.97 (1H, dd, J = 7.5 & 1.0 Hz, H-12), 11.43 (1H, s, HO-1), 12.42 (1H, s, HO-14).

^{13}C NMR (CDCl$_3$, 150 MHz at 25 °C) δ (ppm): 22.7, 36.7, 37.4, 40.8, 56.4, 56.6, 67.5, 67.6, 94.4, 108.6, 114.0, 116.1, 118.4, 119.4, 119.4, 119.7, 121.6, 122.3, 128.3, 128.6, 128.7, 135.0, 135.1, 135.1, 135.5, 136.0, 148.2, 151.5, 160.6, 161.3, 162.2, 164.7, 169.5, 181.9, 188.0.

Protected TMC-66 (2.61)
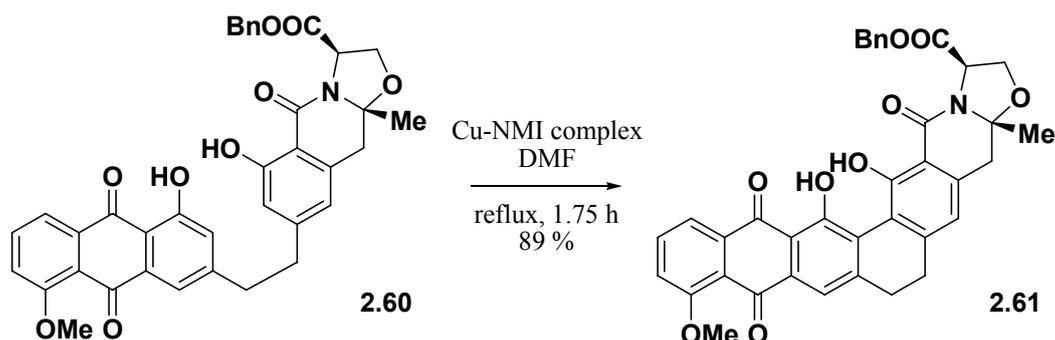
Preparation of copper-N-methylimidazole complex

The following reaction was carried out under an oxygen atmosphere. To a solution of CuCl 496 mg (5.01 µmol) in 95% methanol was added *N*-methylimidazole 1.60 ml (20.0 µmol), and the mixture was stirred at room temperature for 1 hour. The resulting precipitates were collected by filtration, washed with acetone, and dried in vacuo to give the desired copper *N*-methylimidazole complex 1.29 g as a dark green powder.

mp : 149.7-151.3 °C (decomp.)

Synthesis of compound 2.61

The following reaction was carried out under air. To a solution of the compound **2.60** 6.0 mg (9.5 µmol) in DMF 300 µl was added Cu-NMI complex 8.0 mg (29 µmol), and the mixture was heated at 155 °C for 1.75 hours. After cooling to 0 °C, the reaction mixture was quenched with sat. KHSO$_4$ 100 µl and extracted with ethyl acetate (3×300 µl). The combined extracts were evaporated in vacuo. The crude residue was purified by flash column chromatography (toluene-ethyl acetate 3:1) to yield the heptacycle **2.61** as an orange solid (5.3 mg, 8.4 µmol, 89%).

R_f : 0.32 (toluene-ethyl acetate 3:1)

HR-MS (FAB+): calcd. for C$_{37}$H$_{30}$NO$_9$: 632.1921, found 632.1931 [M+H]$^+$.

$[\alpha]_D^{25}$ -115.5 ° (c, 1.08, CHCl$_3$)

mp : 143.4-152.2 °C

IR (cm^{-1}, KBr): 2946, 1749, 1662, 1627, 1272, 750.

^{1}H NMR (CDCl$_3$, 600 MHz at 25 °C) δ (ppm): 1.54 (3H, s, Me-21), 2.61-3.03 (4H, m, H-5 & H-6), 3.11 (1H, d, J = 15.0 Hz, H-19), 3.18 (1H, d, J = 15.0 Hz, H-19), 4.06 (3H, s, OMe), 4.20 (1H, dd, J = 9.0 & 7.0 Hz, H-17), 4.50 (1H, dd, J = 9.0 & 8.0 Hz, H-17), 4.86 (1H, dd, J = 8.0 & 7.0 Hz, H-16), 5.26 (1H, d, J = 12.0 Hz, O-C$\underline{H}_2$-Ph), 5.29 (1H, d, J = 12.0 Hz, O-C$\underline{H}_2$-Ph), 6.68 (1H, s, H-4), 7.35 (1H, dd, J = 8.5 &

1.0 Hz,H-10), 7.32-7.45 (5H, m, Ph), 7.70 (1H, s, H-7), 7.73 (1H, dd, J = 8.5 & 8.0 Hz, H-11), 8.01 (1H, dd, J = 8.0 & 1.0 Hz, H-12), 12.58 (1H, br s, HO-1), 13.48 (1H, br s, HO-14).

^{13}C NMR (CDCl$_3$, 150 MHz at 25 °C) δ (ppm): 22.8, 30.3, 30.7, 40.7, 56.6, 56.7, 67.6, 67.7, 94.2, 109.4, 114.8, 117.8, 118.0, 118.2, 119.5, 121.5, 126.3, 128.3, 128.6, 128.7, 133.4, 135.0, 135.1, 135.8, 136.0, 147.6, 149.6, 159.5, 159.5, 160.4, 165.3, 169.5, 182.0, 188.5.

^{1}H NMR (CDCl$_3$, 600 MHz at 50 °C) δ (ppm): 1.53 (3H, s, Me-21), 2.65-2.95 (4H, m, H-5 & H-6), 3.08 (1H, d, J = 15.0 Hz, H-19), 3.17 (1H, d, J = 15.0 Hz, H-19), 4.04 (3H, s, OMe), 4.20 (1H, dd, J = 9.0 & 6.5 Hz, H-17), 4.47 (1H, dd, J = 9.0 & 8.5 Hz, H-17), 4.86 (1H, dd, J = 8.5 & 6.5 Hz, H-16), 5.26 (1H, d, J = 12.0 Hz, O-C$\underline{H}_2$-Ph), 5.28 (1H, d, J = 12.0 Hz, O-C$\underline{H}_2$-Ph), 6.65 (1H, s, H-4), 7.33 (1H, dd, J = 8.5 & 1.0 Hz,H-10), 7.30-7.43 (5H, m, Ph), 7.68 (1H, s, H-7), 7.71 (1H, dd, J = 8.5 & 7.5 Hz, H-11), 8.01 (1H, dd, J = 7.5 & 1.0 Hz, H-12), 12.46 (1H, br s, HO-1), 13.36 (1H, br s, HO-14).

^{13}C NMR (CDCl$_3$, 150 MHz at 50 °C) δ (ppm): 23.0, 30.4, 30.8, 40.9, 56.6, 56.8, 67.7, 67.7, 94.3, 109.6, 114.9, 117.7, 117.9, 118.5, 119.2, 119.6, 121.9, 126.5, 128.3, 128.5, 128.7, 133.6, 134.9, 135.3, 136.0, 136.1, 147.6, 149.6, 159.4, 159.7, 160.5, 165.3, 169.5, 181.9, 188.5.

TMC-66 (**2.1**)

2.61   BBr$_3$ CH$_2$Cl$_2$ -78 °C, 30 min   70 %   **TMC-66 (2.1)**
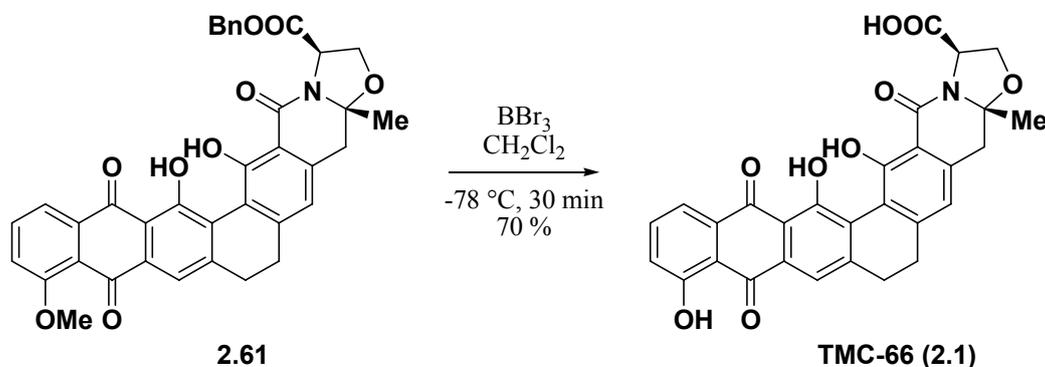
To a cooled (-78 °C) solution of the compound **2.61** 5.3mg (8.4 μmol) in CH$_2$Cl$_2$ 650 μl was added boron tribromide (1.0 M in CH$_2$Cl$_2$, 42.0 μl, 42 μmol) and the mixture was stirred at -78 °C for 30 minutes. The reaction mixture was quenched with sat. NaHCO$_3$ 100 μl and stirred at 0 °C for 30 minutes. The mixture was added sat. KHSO$_4$ 200 μl and the mixture was extracted with EtOAc (2×650 μl). The combined extracts were evaporated in vacuo. The crude residue was purified by flash column chromatography (toluene-ethyl acetate 3:1 0.5% aq. TFA) to yield the TMC-66 (**2.1**) as ared solid(3.1 mg, 5.9 μmol, 70%)

R_f : 0.50 (CHCl$_3$-MeOH 3:1)
HR-MS (FAB+): calcd. for C$_{29}$H$_{22}$NO$_9$: 528.1295, found 528.1289 [M+H]$^+$.
[α]$_D^{26}$ -60.0 ° (c, 0.01, CHCl$_3$), [α]$_D^{26}$ -73.3 ° (c, 0.09, CHCl$_3$)
mp : 215-220 °C (dec.)
IR (cm^{-1}, KBr): 3446, 2950, 1733, 1616, 1270, 754.
^{1}H NMR (CDCl$_3$, 600 MHz at 25 °C) δ (ppm): 1.52 (3H, s, Me-21), 2.59-3.08 (4H, m, H-5 & H-6), 3.17 (1H, d, J = 15.5 Hz, H-19), 3.25 (1H, d, J = 15.5 Hz, H-19), 4.56 (2H, d, J = 8.0 Hz, H-17), 4.88 (1H, t, J = 8.0 Hz, H-16), 6.73 (1H, s, H-4), 7.31 (1H, dd, J = 8.5 & 1.0 Hz,H-10), 7.68 (1H, dd, J = 8.5 & 7.5 Hz, H-11), 7.77 (1H, s, H-7), 7.87 (1H, dd, J = 7.5 & 1.0 Hz, H-12), 12.15 (1H, br s, HO-1), 12.68 (1H, s, HO-9), 13.68 (1H, br s, HO-14).

^{13}C NMR (CDCl$_3$, 150 MHz at 25 °C) δ (ppm): 22.7, 30.2, 30.4, 40.6, 56.7, 66.3, 94.2, 108.6, 115.2, 116.1, 117.9, 118.2, 119.2, 119.4, 124.7, 128.0, 131.5, 133.5, 136.5, 136.7, 148.7, 149.3, 159.4, 160.3, 162.6, 166.7, 170.3, 187.9, 187.9.

¹H NMR (DMSO-*d*₆, 600 MHz at 50 °C) δ (ppm): 1.43 (3H, s, Me-21), 2.55-3.11 (4H, m, H-5 & H-6), 3.18 (1H, d, *J* = 15.0 Hz, H-19), 3.25 (1H, d, *J* = 15.0 Hz, H-19), 4.15 (1H, dd, *J* = 9.0 & 7.0 Hz, H-17), 4.56 (1H, dd, *J* = 9.0 & 8.5 Hz, H-17), 4.71 (1H, dd, *J* = 8.5 & 7.5 Hz, H-16), 6.87 (1H, s, H-4), 7.40 (1H, dd, *J* = 8.0 & 1.0 Hz,H-10), 7.73 (1H, s, H-7), 7.78 (1H, dd, *J* = 7.5 & 1.0 Hz, H-12), 7.83 (1H, dd, *J* = 8.0 & 7.5 Hz, H-11), 12.52 (1H, s, HO-9), 12.81 (1H, s, HO-1 or HO-14), 13.37 (1H, s, HO-1 or HO-14).
¹³C NMR (DMSO-*d*₆, 150 MHz at 50 °C) δ (ppm): 22.4, 29.1, 29.6, 39.9, 56.3, 67.2, 93.5, 108.8, 114.5, 115.8, 117.6, 117.8, 117.8, 118.9, 124.7, 127.7, 130.8, 133.1, 137.0, 137.1, 147.3, 149.5, 158.3, 159.3, 161.5, 164.2, 170.8, 187.3, 187.4.
¹H NMR (DMSO-*d*₆, 600 MHz at 90 °C) δ (ppm): 1.45 (3H, s, Me-21), 2.65-3.00 (4H, m, H-5 & H-6), 3.17 (2H, s, H-19), 4.16 (1H, dd, *J* = 8.5 & 6.0 Hz, H-17), 4.50 (1H, dd, *J* = 8.5 & 8.0 Hz, H-17), 4.72 (1H, dd, *J* = 8.0 & 6.0 Hz, H-16), 6.82 (1H, s, H-4), 7.36 (1H, dd, *J* = 6.5 & 3.0 Hz,H-10), 7.72 (1H, s, H-7), 7.77-7.83 (2H, m, H-11 & H-12)
¹³C NMR (DMSO-*d*₆, 150 MHz at 90 °C) δ (ppm): 22.4, 29.1, 29.6, 39.9, 56.3, 67.2, 93.5, 108.8, 114.5, 115.8, 117.6, 117.8, 117.8, 118.9, 124.4, 127.9, 130.8, 133.1, 137.0, 137.1, 147.3, 149.5, 158.4, 159.3, 161.5, 164.2, 170.8, 187.3, 187.4.

References

1) K. Shindo, H. Kawai, *J. Antibiot.* **1992**, *45*, 584-586.

2) N. Naruse, M. Goto, Y. Watanabe, T. Terasawa, K. Dobashi, *J. Antibiot.* **1998**, *51*, 545-552.

3) O. Potterat, H. Zähner, C. Volkmann, A. Zeeck, *J. Antibiot.* **1993**, *46*, 346-349.

4) T. Oja, K. Palmu, H. Lehmussola, O. Leppäranta, K. Hännikäinen, J. Niemi, P. Mäntsälä, M. Mestä-Ketelä, *Chemistry & Biology,* **2008**, *15*, 1046-1057.

5) (a) K. Tatsuta, K. Akimoto, M. Annaka, Y. Ohno, M. Kinoshita, *J. Antibiot.* **1985**, *38*, 680-682.; (b) K. Tatsuta, K. Akimoto, M. Annaka, Y. Ohno, M. Kinoshita, *Bull. Chem. Soc. Jpn.*, **1985**, *58*, 1699-1706.

6) K. Tatsuta, H. Ozeki, M. Yamaguchi, M. Tanaka, T. Okui, *Tetrahedron Lett.* **1990**, *31*, 5495-5498.

7) K. Tatsuta, T. Hirabayashi, M. Kojima, Y. Suzuki, T. Ogura, *J. Antibiot.* **2004**, *57*, 291-297.

8) K. Tatsuta, Y. Suzuki, T. Toriumi, Y. Furuya, S. Hosokawa, *Tetrahedron Lett.* **2007**, *48*, 8018-8021.

9) K. Tsukamoto, M. Hirayama, S. Nakajima, T. Okabe, K. Kojiri, H. Suda, Kokai Tokkyo Koho, **1997**, JP9-241257.

10) B. Bieber, J. Nüske, M. Ritzau, U. Grafë, *J. Antibiot.* **1998**, *51*, 381-382.

11) Björn, B., Jörg, N., German patent, **1998**, DE19745914.

12) (a) P. Jarglis, F. W. Lichtenthaler, *Tetrahedron Lett.* **1982**, *23*, 3781-3784,; (b) K. Tatsuta, T. Yamazaki, T. Mase, T. Yoshimoto, *Tetrahedron Lett.* **1998**, *39*, 1771-1772,; (c) M. Carda, S. Rodríguez, F. González, E. Castillo, A. Villanueva, J. A. Marco, *Eur. J. Org. Chem.* **2002**, 2649-2655.

13) (a) D. J. Dodsworth, M. Calcagno, E. U. Ehrmann, B. Devadas, P. G. Sammes, *J. Chem. Soc., Perkin Trans.* 1, **1981**, 2120-2124,; (b) M. Wierzbicki, M. Boussard, A. Rousseau, J. A. Boutin, P. Delagrange, *United States Patent*, **2002**, US6465660B1,; (c) C.Cox, S. J. Danishefsky, *Org. Lett.* **2000**, *2*, 3493-3496.

14) Y. Asai, N. Nonaka, S. Suzuki, M. Nishio, K. Takahashi, H. Shima, K. Ohmori, T. Ohnuki, S. Komatsubara, *J. Antibiot.* **1999**, *52*, 607-612.

15) McMahon, E. G.; M. A. Palomo, W. M. Moore, J. F. Mcdonald, M. K. Stern, Proc. Natl. Acad. Sci. USA 88: 703-707, **1991**.

16) Y. Tsurumi, N. Ohhata, T. Iwamoto, N. Shigematsu, K. Sakamoto, M. Nishikawa, S. Kiyoto, M. Okuhara, *J. Antibiot.* **1994**, *47*, 619-630.

17) M. Ritzau, R. Vettermann, W. F. Fleck, W. Gutsche, K. Dornberger, U. Gräfe, *J. Antibiot.* **1997**, *50*, 791-793.

18) (a) H. Umezawa, T. Takeuchi, T. Hara, H. Naganawa, M. Okada, M. Hamada, S. Gomi, M. Sezaki, S. Kondo, *J. Antibiot.* **1988**, *41*, 807-811,; (b) T. Oki, M. Konishi, K. Tomatsu, K. Tomita, K. Saitoh, M. Tsunakawa, M. Nishio, T. Miyaki, H. Kawaguchi, *J. Antibiot.* **1988**, *41*, 1701-1704.

19) A. Nakagawa, S. Omura, K. Kushida, H. Shimizu, G. Luckacs, *J. Antibiot.* **1987**, *40*, 301-308.

20) (a) T. Nishizuka, S. Hirosawa, S. Kondo, D. Ikeda, T. Takeuchi, *J. Antibiot.* **1997**, *50*, 755-764,; (b)

K. Krohn, S. Bernhard, U. Flörke, N. Hayat, *J. Org. Chem.* **2000**, *65*, 3218-3222,; (c) F. M. Hauser, H. Liao, Y. Sun, *Org. Lett.* **2002**, *4*, 2241-2243,; (d) T. R. Kelly, W. Xu, Z. Ma, Q. Li, V. Bhushan, *J. Am. Chem. Soc.* **1993**, *115*, 5843-5844,; (e) T. R. Kelly, Q. Li, V. Bhushan, *Tetrahedron Lett.* **1990**, *31*, 161-164,; (e) M. Kitamura, K. Ohmori, T. Tawase, K. Suzuki, *Angew. Chem.* **1999**, *111*, 1308-1311; *Angew. Chem. Int. Ed.* **1999**, *38*, 1229-1232,; (f) K. Ohmori, M. Tamiya, M. Kitamura, H. Kato, M. Oorui, K. Suzuki, *Angew. Chem.* **2005**, *117*, 3939-3942; *Angew. Chem. Int. Ed.* **2005**, *44*, 3871-3874.

21) (a) T. R. Kelly, C. T. Jagoe, Q. Li, *J. Am. Chem. Soc.* **1989**, *111*, 4522-4524,; (b) A. V. R. Rao, J.S. Yadav, K. K. Reddy, V. Upender, *Tetrahedron Lett.* **1991**, *32*, 5199-5202,; (c) G. Mehta, S. R. Shah, Y. Venkateswarlu, *Tetrahedron,* **1994**, *50*, 11729-11742.

22) (a) M. Kosugi, I. Hagiwara, T. Migita, *ChemLett.*, **1983**, 839,; (b) M. Kosugi, I. Hagiwara, T. Sumiya, T. Migita, *Bull. Chem. Soc. Jpn.* **1984**, *57*, 242.

23) W. J. Scott, G. T. Crisp, J. K. Stille, *J. Am. Chem. Soc.* **1984**, *106*, 4630.

24) (a) J. M. Fox, X. Huang, A. Chieffi, S. L. Buchwald, *J. Am. Chem. Soc.* **2000**, *122*, 1360,; (b) O. Gaertzen, S. L. Buchwald, *J. Org. Chem.* **2002**, *67*, 465,; (c) P. Liu, T. J. Lanza, Jr., J. P. Jewell, C. P. Jones, W. K. Hagmann, L. S. Lin, *Tetrahedron Lett.* **2003**, *44*, 8869.

25) J. Hassan, M. Sévignon, C. Gozzi, E. Schulz, M. Lemaire, *Chem. Rev.* **2002**, *102*, 1359.

26) M. Noji, M. Nakajima, K.Koga, *Tetrahedron Lett.* **1994**, *35*, 7983.

27) R. Fulwood, D. Parker, *J. Chem. Soc., Perkin Trans. 2*, **1994**, 57.

Acknowledgements

Achievements for this dissertation

1) The first total synthesis and structural determination of antibiotics K1115 B$_1$s (alnumycins)

Tetrahedron Lett., in press.

Kuniaki Tatsuta, Sonoko Tokishita, <u>Tomohiro Fukuda</u>, Takaaki Kano, Tadaaki Komiya, and Seijiro Hosokawa

2) The first total synthesis and structural determination of TMC-66

Tetrahedron Lett. **2007**, *48*, 7305-7308.

Seijiro Hosokawa, Hitoshi Fumiyama, Hisato Fukuda, <u>Tomohiro Fukuda</u>, Masashi Seki, and Kuniaki Tatsuta

3) Total synthesis of an endothelin converting enzyme inhibitor, TMC-66

Heterocycles, **2008**, *76*, 699-713.

Seijiro Hosokawa, Hitoshi Fumiyama, Hisato Fukuda, <u>Tomohiro Fukuda</u>, Masashi Seki, and Kuniaki Tatsuta