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ORIGINAL ARTICLE

Toward the total synthesis of luminamicin; an anaerobic antibiotic: construction of highly functionalized *cis*-decalin containing a bridged ether moiety

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Synthesis of a *cis*-decalin moiety, containing an oxa-bridged *cis*-decalin ring system (11-oxatricyclo(5.3.1.^{1,7}0^{3,8})undecane), as a key intermediate of the total synthesis of luminamicin (1) was accomplished. One of the essential steps in our synthetic route is construction of a *cis*-decaline framework using a one-pot Michael addition-aldol reaction. Additionally, the bridged ether moiety was obtained by an intramolecular 1,6-oxa-Michael reaction of a conjugated aldehyde.
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INTRODUCTION

Pseudomembranous colitis, caused by overgrowth of *Clostridium difficile*, is often related to recent antibiotic consumption. The condition is increasing and regarded as a serious problem around the world.[1,2] There are few antibacterial drugs (vancomycin, metronidazole, and so on) effective against anaerobic bacteria and there is a growing concern about the emergence and spread of drug-resistant bacteria and ineffective treatment. We have focused on the screening of compounds to combat anaerobic bacteria from microbial metabolites, and a natural product luminamicin (**1**) was found from the fermentation broth of *Streptomyces* sp. OMR-59 in 1985.[3] Notably, **1** shows selective activity against *Clostridium* sp., with a minimum inhibitory concentration (MIC) value of 1.0 μg ml^{-1}. Two years after its discovery, McAlpin and co-workers isolated coloradocin, and recognized it was the same compound as **1** based on spectral data analysis.[4,5] Additionally, they determined the relative structure of **1** (Figure 1). It possesses several interesting structural features, including a multi-functional oxa-bridged *cis*-decalin ring system, with a 10-membered and a 14-membered macrolactone with an enol ether conjugated with a maleic anhydride moiety. However, it proved inactive against most aerobic bacteria. In contrast, two structurally related natural products, nodusmicin[6] and nargenicin,[7] do exhibit antibacterial activity against some anaerobic bacteria.[8]

We were therefore interested in studying the key structure moieties responsible for the specific anti-anaerobic bacteria activity. Thus, we determined the absolute stereostructure of **1** using conformational analysis via high-temperature dynamics, NMR spectroscopy, and the modified Mosher method,[9] and devised a route to synthesize the compound with an expectation of discovering lead compounds for the development of novel drugs to overcome anaerobic bacteria.

We have previously reported synthesis of the 14-membered macrolactone framework containing conjugated maleic anhydride as one of the characteristic structures in **1**.[10] The next synthetic challenge for the total synthesis of **1** was construction of the oxa-bridged *cis*-decalin system. Kallmerten's group described the total synthesis of (+)-deoxynargenicin[11] and Mulzer's group reported the total synthesis of branimycin.[12–14] Both natural products are structurally similar to **1**. In their reports, the racemic *cis*-decaline moiety of branimycin was synthesized by an intramolecular Diels–Alder reaction, and the bridged ether moiety was constructed using regioselective opening of epoxide by alkoxide, which has a different oxa-bridge system from that of **1**. We have previously developed an efficient route for synthesizing 11-oxatricyclo[5.3.1.^{1,7}0^{3,8}) undecane,[15] but further elaboration was difficult at the C-4, 10 and 11 positions. Therefore, a new construction method of oxa-bridged *cis*-decalin is needed to complete the total synthesis of **1**, and herein we report a stereo-controlled synthetic route to generate the oxa-bridged *cis*-decalin ring unit (**2**).

RESULTS AND DISCUSSION

Our synthetic approach is shown in Scheme 1. The highly-strained tricyclic compound **2** would be obtained by stereoselective protonation of conjugated silyl enol ether (**3**). Oxa-bridged moiety of **3** would

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Dedicated to Professor KC Nicolaou and his outstanding contributions to complex natural product total synthesis and chemical biology.
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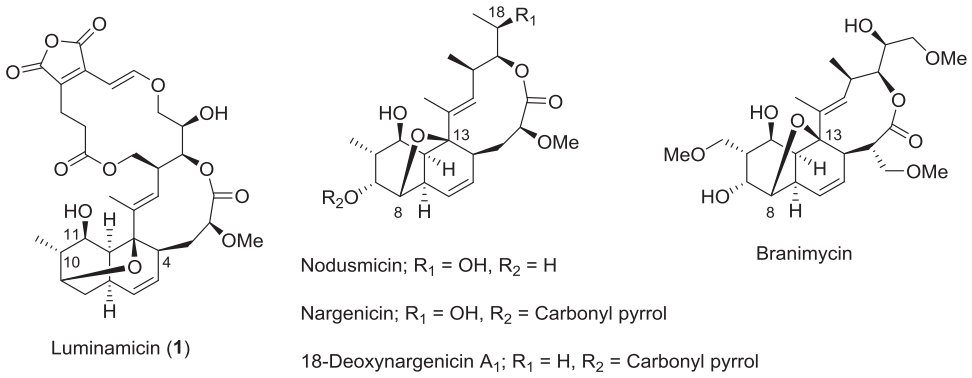
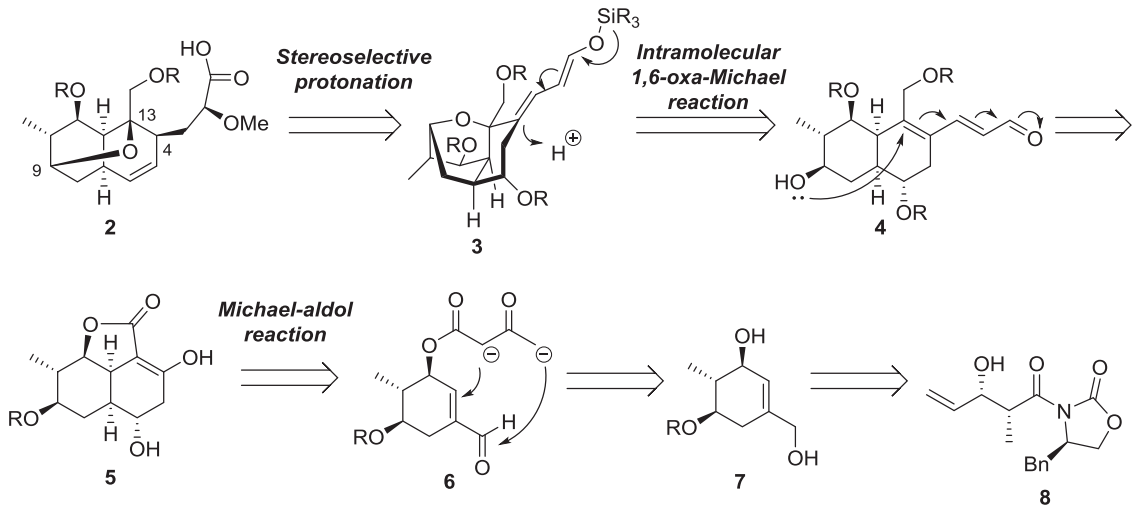


Figure 1 Structures of luminamicin (**1**) and structurally related natural products.



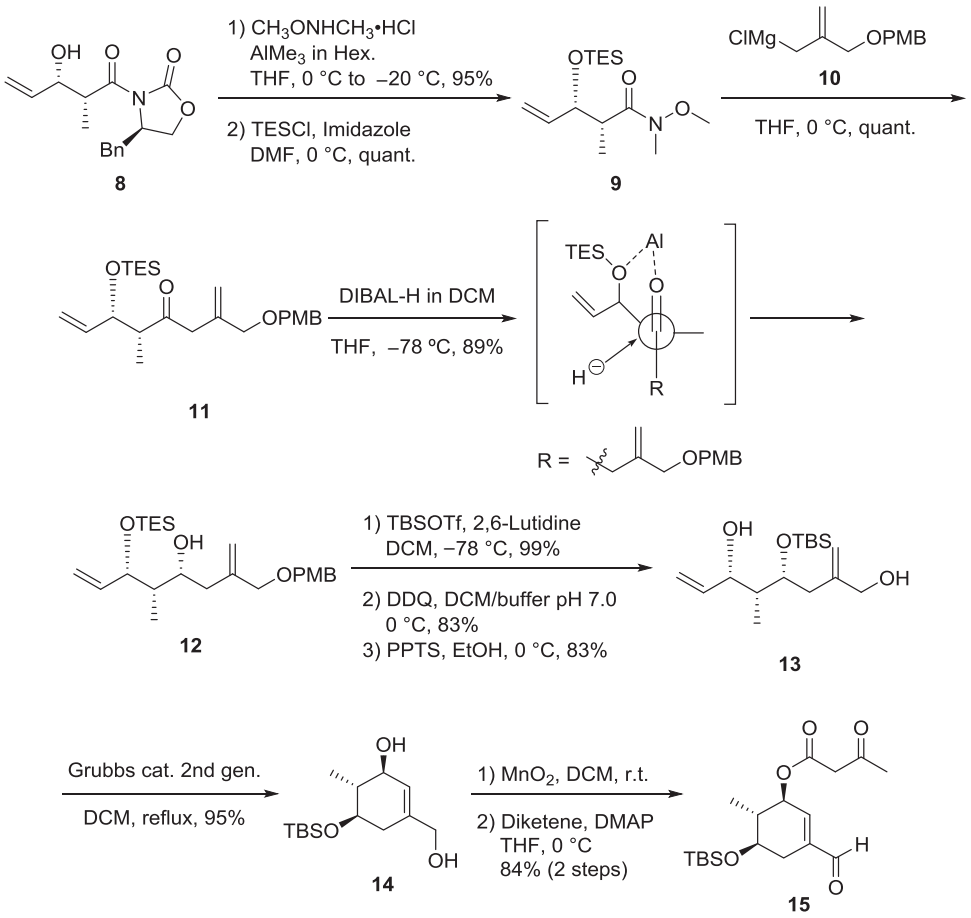
Scheme 1 Retrosynthetic analysis of the key intermediate **2**.

be constructed via an intramolecular 1,6-oxa-Michael reaction of the conjugated aldehyde (**4**), which could be obtained from the tricyclic compound (**5**) by introduction of a side chain and reduction of the lactone moiety. *cis*-Decalin **5** could be constructed by chiral transferring Michael–aldol cascade reaction of the α,β-unsaturated aldehyde (**6**). Optically pure tri-substituted cyclohexene (**7**) could be prepared from known Evans aldol product (**8**)[16,17] via stereoselective reduction of ketone and ring-closing metathesis (RCM).

Our first task was preparation of optically pure **7** for construction of the *cis*-decalin framework. The *syn*-aldol product **8**, available in chiral form using the protocol reported by Evans, was converted to the Weinreb amide, followed by TES protection to afford the amide (**9**) in 95% yield over two steps (Scheme 2). Subsequently, **9** was treated with the Grignard reagent,[18] to produce the ketone (**11**). We next performed stereoselective reduction of the ketone, when using DIBAL-H, the high diastereoselectivity was attributed to a six-membered ring transition state with the hydride, eventually and obtaining alcohol (**12**) (*d.r.* = 20:1). Notably, when **11** was treated with $NaBH_4$, desired alcohol and its diastereomer was obtained (*d.r.* = 3:1). The newly generated stereocenter was determined by Rychnovsky's protocol [19] to be the desired (*R*) configuration. The secondary alcohol was protected as the TBS ether, and removal of the PMB and TES groups, afforded the bis alcohol (**13**). Ring-closing metathesis of **13** gave the cyclohexene (**14**), and subsequent oxidation of the primary alcohol and acylation afforded the required cascade cyclization precursor (**15**).

The Michael-aldol cascade reaction was one of the most challenging steps in our synthesis strategy. Previously, Kitahara *et al.*[20] reported a similar one-pot reaction to synthesize (+)-tetrahydroisocoumarin.[20] In their report, 1*H*-2-benzopyran-1-one was constructed via a one-pot esterification-Michael addition-aldol reaction using Cs_2CO_3. As the first trial, we treated **15** with Cs_2CO_3 in benzene reflux condition but only decomposition of substrate was observed. On the basis of this result, using milder base reagent K_2CO_3 under the same condition, gave the desired tricyclic product (**16**) in moderate yield. To further improve the yield, we performed the Michael addition at 60 °C first and then the aldol reaction at reflux in toluene, which gave the Michael-aldol product in 82% yield without decomposition and with good reproducibility at a multi-gram scale (Scheme 3).

Preparation of vinyl triflate using Tf_2O, Et_3N condition, followed by Stille coupling with **16** and (*E*)-vinylstannane (**17**),[21] provided conjugated γ-lactone (**18**) in excellent yield. To improve the yield, we used polar aprotic DMF as the solvent. After protection of the secondary alcohol as the BOM ether, reduction of lactone with $LiAlH_4$



Scheme 2 Synthesis of the cascade cyclization precursor **15**.

afforded the diol. Selective protection of the primary alcohol using AcCl, collidine, under − 55 °C condition, followed by protection of the secondary alcohol produced the MEM ether (**20**). Our second key operation was construction of the highly-rigid tricyclic compound. Deprotection of the TBS group gave the secondary alcohol. Subsequent removal of the PMB group simultaneously oxidized the allyl alcohol to provide the precursor conjugated aldehyde (**21**) as the 1,6-oxa-Michael reaction precursor.

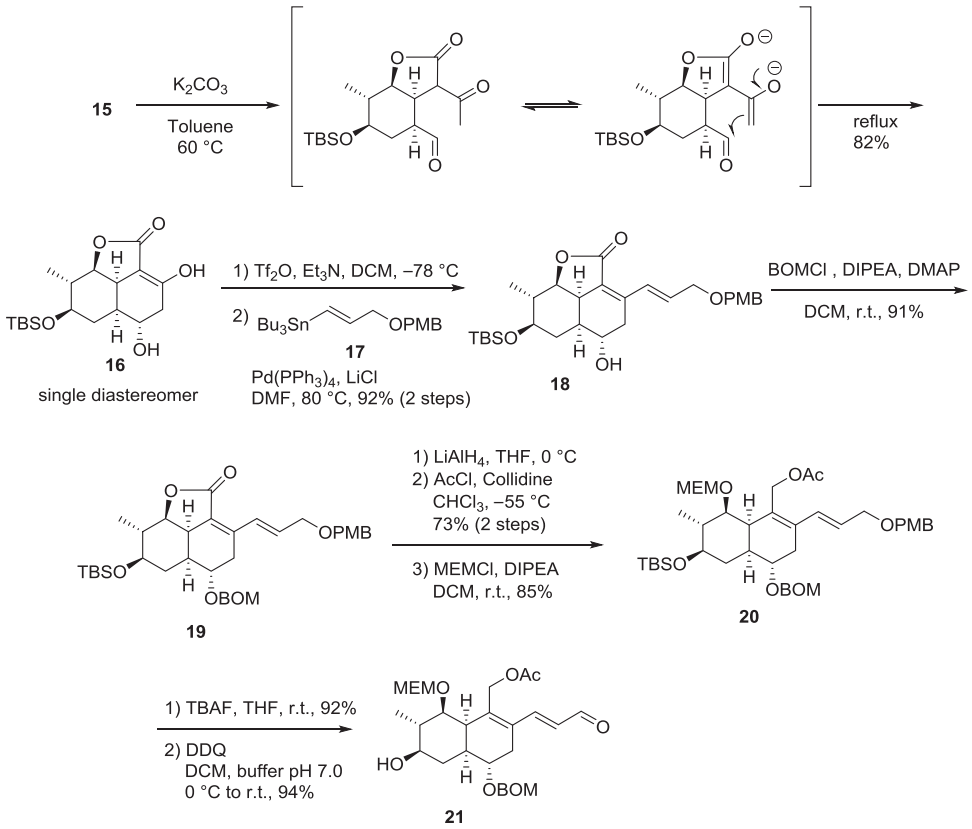
We next attempted construction of the oxa-bridge with an intramolecular 1,6-oxa-Michael reaction being a key component. Since the desired product formed boat conformation, we were concerned about a retro-Michael reaction. Therefore, activation of the aldehyde had to be made, along with *in situ*-generated enolate trap, as the silyl enol ether. When **21** was treated with TBSOTf, we obtained the silylated secondary alcohol instead of the silyl enol ether. Therefore, we changed the Lewis acid to TIPSOTf, a more bulky one, to avoid the undesired silylation on secondary alcohol. This change of the reaction condition gave the silyl enol ether (**22**) in 99% yield (Scheme 4). With the oxa-bridged compound in hand, we investigated the stereoselective protonation of the C-4 position toward construction of **2**. It was anticipated that we could encounter retro-oxa-Michael reaction in this step. As our first attempt, TBAF mainly resulted in retro-oxa-Michael reaction in moderate yield. To suppress this side reaction, TASF was used under neutral condition, providing the desired product and C-4-oxidized product (1.75:1). Dibutylhydroxytoluene (BHT) was added to the reaction condition to trap the oxy radical in degassed THF, oxidative product was not generated and **23** was obtained as a major product. Stereochemistry of **23** at C-4 position was determined by ^{1}H NMR (*c.a.* 9:1) and nuclear Overhauser effect (nOe) analyses of saturated methyl ester (**25**). The conjugated aldehyde **23** was used in the next reaction without further purification, considering its instability. After converting **23** to the methyl ester **24**, hydrogenation of α,β-unsaturated ester and removal of the BOM group was accomplished using Raney nickel. Subsequent monochloromesylation and dehydration gave **25** as a single diastereomer after purification in 48% yield over five steps. At this stage, the key intermediate having all the stereocenters contained in *cis*-decalin was synthesized.

CONCLUSION

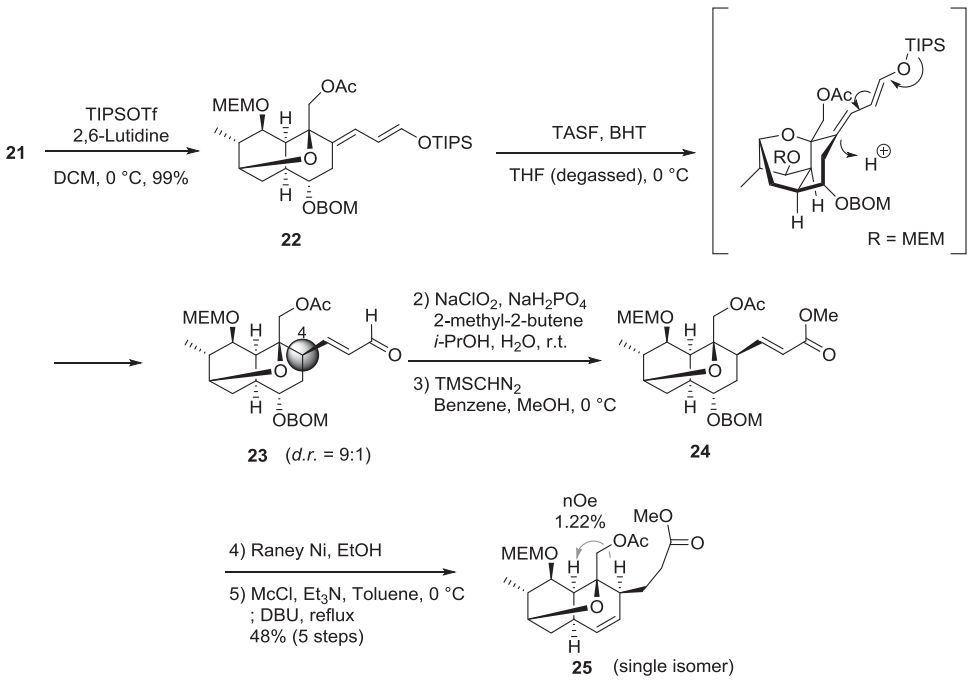
In conclusion, we have achieved the synthesis of an essential intermediate, including an oxa-bridged *cis*-decalin ring system (11-oxatricyclo(5.3.1.^{1,7}0^{3,8})undecane) necessary for the total synthesis of **1**. To obtain the essential framework, we exploited a stereocontroled Michael-aldol cascade reaction plus an intramolecular 1,6-oxa-Michael reaction to achieve the construction of the key intermediate **25**. Further studies toward the total synthesis of **1** are now in progress.

MATERIALS AND METHODS

Detail of experimental procedures, characterization data and NMR spectra for all new compounds can be found in Supplementary Information.



Scheme 3 Synthesis of conjugated aldehyde **21**.



Scheme 4 Synthesis of key intermediate **25** for total synthesis of **1**.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

ACKNOWLEDGEMENTS

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Supplementary Information accompanies the paper on The Journal of Antibiotics website (http://www.nature.com/ja)