

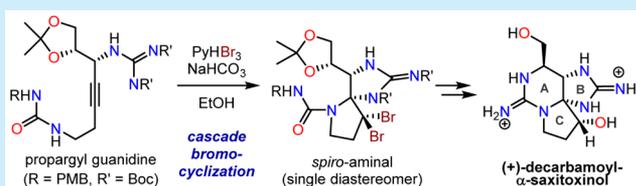
# A Synthetic Strategy for Saxitoxin Skeleton by a Cascade Bromocyclization: Total Synthesis of (+)-Decarbamoyl- $\alpha$ -saxitoxinol

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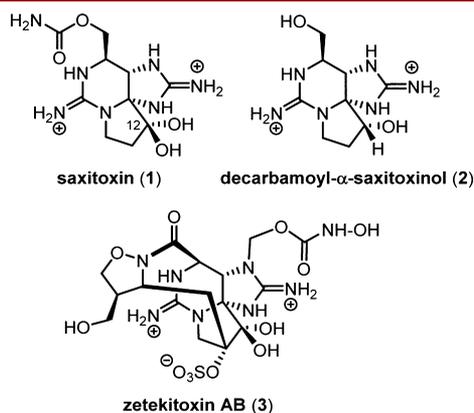
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**S** Supporting Information

**ABSTRACT:** A new synthetic strategy for the formation of the ABC tricyclic framework of saxitoxin was developed. The BC ring moiety, including a *spiro*-aminal structure, was first constructed stereoselectively by a newly designed cascade bromocyclization of a readily available internal alkyne bearing guanidine and urea. The A ring was then synthesized by a guanylation of a cyclic urea, easily prepared via the oxidative cleavage of the diol of the cascade product, followed by addition of cyanide. This strategy enables the concise stereocontrolled total synthesis of (+)-decarbamoyl- $\alpha$ -saxitoxinol, which is a naturally occurring saxitoxin analogue.



Saxitoxin (STX, **1**; Figure 1), a paralytic shellfish poison (PSP),<sup>1</sup> is one of the best-known marine toxins along with



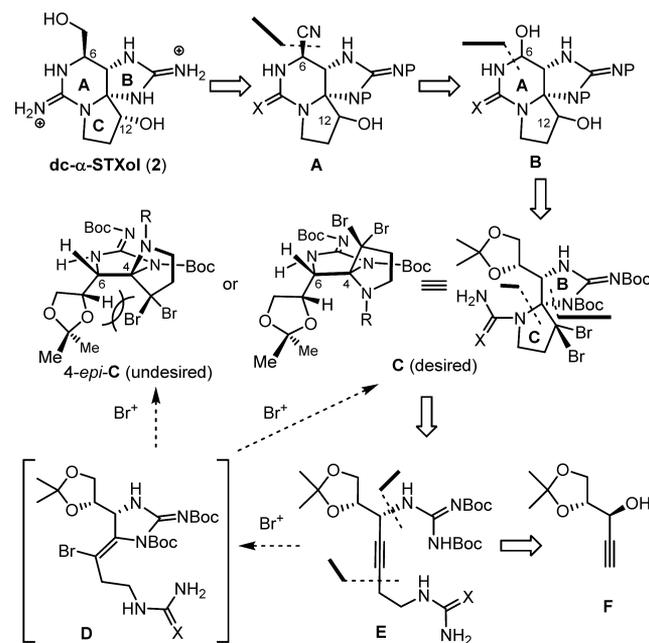
**Figure 1.** Chemical structures of saxitoxin and its analogues.

tetrodotoxin (TTX), i.e., the puffer fish toxin. Since these two guanidine-containing natural products exhibit potent and highly selective inhibitory activity toward voltage-gated sodium channels (VGSC), both have found widespread use as indispensable biochemical tools in physiological experiments associated with ion channels.<sup>2</sup> Recently, STX has been featured as a scaffold in the development of subtype-selective inhibitors of VGSCs, which are expected to be useful in the analysis of the functions of individual subtypes of VGSCs.<sup>3</sup> In order to prepare diverse STX analogues, including naturally occurring neoSTX, GTX, and zetekitoxin AB (**3**), efficient synthetic routes for a range of STX analogues have been developed.<sup>4–8</sup> We have also developed a synthetic strategy for the generation of the STX skeleton via a  $\text{Br}^+$ -triggered cascade cyclization of guanidino acetylene, which may find applications in the exploration of subtype-selective blockers of VGSCs.<sup>9</sup> Herein, we describe an

efficient alternative strategy toward the STX skeleton, in which a different cascade bromocyclization reaction is a key step.

The newly designed synthetic strategy for the STX skeleton is shown in Scheme 1 and exemplified by the synthesis of the naturally occurring STX analogue decarbamoyl- $\alpha$ -saxitoxinol (dc- $\alpha$ -STXol, **2**).<sup>10</sup> We envisioned that **2** could be prepared from intermediate **A** ( $\text{X} = \text{NH}_2$ : guanidine, or  $\text{X} = \text{O}$ : urea) by transforming the nitrile into a primary alcohol and by

## Scheme 1. Synthetic Plan for the Formation of the STX Skeleton



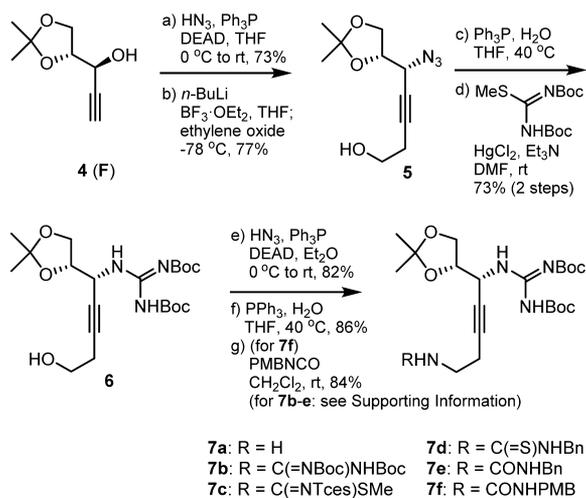
**Received:** October 31, 2016

guanylation of urea. Since the nitrile is synthetically equivalent to a variety of functional groups, intermediate **A** might be used as a common intermediate for the synthesis of diverse STX analogues that differ at the C-6 position. The nitrile moiety could be introduced stereoselectively by addition of cyanide to the *N,O*-acetal of **B** from the convex face of the AB ring. Compound **B** could be easily prepared from **C** including a *spiro*-aminal structure, whose structure has not been utilized before because *spiro*-aminals are difficult to prepare in general;<sup>11</sup> the *N,O*-acetal should be prepared by cleavage of the 1,2-glycol moiety of **C**, while the hydroxy group at the C-12 position could be prepared from *gem*-dibromomethylene via a previously developed radical-mediated oxygenation.<sup>12</sup>

We envisaged that the *spiro*-aminal structure of **C** could be constructed in a single step by a Br<sup>+</sup>-triggered cascade cyclization from alkyne **E**, which bears guanidine at the propargyl position, and guanidine (X = NH<sub>2</sub>) or its equivalent (e.g., X = O) at the end of the side chain. The cascade reaction would proceed via intermediate **D** generated by the first bromocyclization of **E**. We anticipated that the second cyclization of **D** proceeds through a late transition state, furnishing the desired diastereomer **C**, because the undesired diastereomer 4-*epi*-**C** exhibits severe steric repulsion between the incoming nitrogen nucleophile and the acetonide-containing side chain. As the nucleophilicity and steric hindrance of nitrogen nucleophiles are strongly influenced by the protective groups or substituents, a screening of the substituents of the amino function and of the reaction conditions would be necessary. Precursor **E** could be prepared from the known propargyl alcohol **F**<sup>13</sup> in several steps. With this analysis in mind, we initially investigated the cascade cyclization of precursor **E**.

Precursors **E** (**7b–f**) were synthesized from known chiral acetylene **4** (**F** in Scheme 1), which was readily prepared from *D*-arabitol in three steps (Scheme 2).<sup>13</sup> The introduction of an

Scheme 2. Synthesis of **7b–f** for the Cascade Cyclization



azide group at the propargyl position of **4** was achieved using Mitsunobu conditions (N<sub>3</sub>H/Ph<sub>3</sub>P, DEAD). Subsequently, the terminal acetylene was hydroxyethylated by the addition of lithium acetylide to ethylene oxide in the presence of BF<sub>3</sub>·OEt<sub>2</sub><sup>14</sup> to afford **5** in 77% yield. The reduction of the azide moiety of **5** under Staudinger conditions (Ph<sub>3</sub>P, THF–H<sub>2</sub>O) was followed by guanylation with di-Boc-*S*-methylisothiourea in

the presence of HgCl<sub>2</sub> and Et<sub>3</sub>N to afford **6** in 73% yield over two steps. The primary alcohol in **6** was transformed into an amine in two steps by the aforementioned reaction sequence. The resulting amine **7a** was used for the preparation of precursors bearing guanidine and its equivalents, e.g., isothiourea, thiourea, and ureas **7b–f**.<sup>15</sup> With these precursors in hand, we investigated the Br<sup>+</sup>-induced cascade cyclization reaction for the construction of *spiro*-aminal **C**.

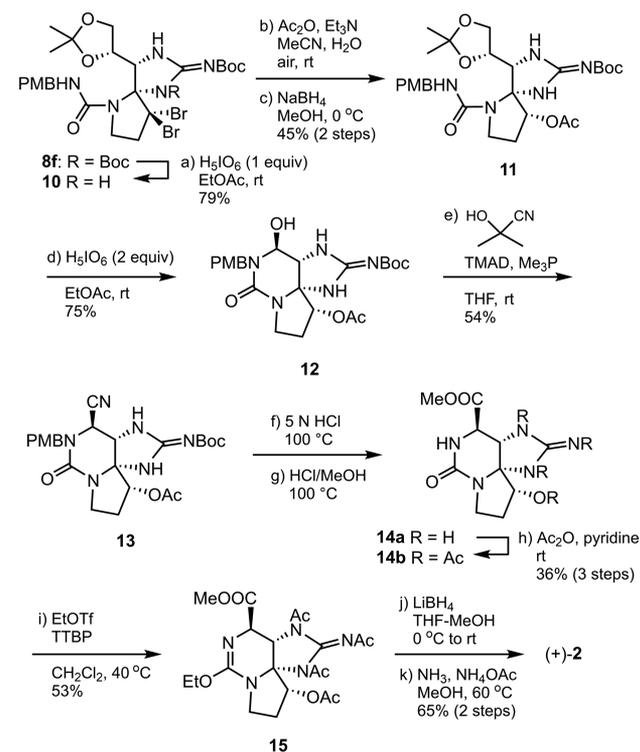
Table 1 shows the results of typical experiments for the development of the cascade bromocyclization using **7b–f**. We initially attempted the cascade cyclization of **7b**, bearing diBoc-guanidine at the terminus of the side chain, using the previously optimized conditions for the synthesis of *spiro*-*N,O*-acetals.<sup>9,16</sup>

When **7b** was treated with 1.3 equiv of pyridinium tribromide (PyHBr<sub>3</sub>) in the presence of K<sub>2</sub>CO<sub>3</sub> in CH<sub>2</sub>Cl<sub>2</sub>/H<sub>2</sub>O, monocyclic guanidine **9b**<sup>17</sup> was obtained in 66% yield (entry 1). However, **9b** was not consumed, even with an excess of PyHBr<sub>3</sub> (entry 2).<sup>18</sup> The reaction of *S*-methyl-isothiourea **7c** with 1.3 equiv of PyHBr<sub>3</sub> also afforded the corresponding monocyclized product **9c** in good yield (entry 3). However, the use of an excess of the reagent (3 equiv) led to the decomposition of **9c** and furnished a complex mixture of products (entry 4), which is probably due to the oxidation of the methyl sulfide moiety in **9c**. In the reaction of thiourea **7d** with PyHBr<sub>3</sub>, the desired product **8a** and monocyclized product **9d** were not detected (entry 5), suggesting facile oxidation of the thiourea under these conditions. Conversely, when benzyl urea **7e** was treated with an excess of PyHBr<sub>3</sub>, **8e** (23%) was observed for the first time together with **9e** (14%) (entry 6). As expected, the desired *R* configuration for the *spiro* center of **8e** was confirmed by single-crystal X-ray diffraction analysis.<sup>19</sup> This result encouraged us to explore the conditions for the use of **7e** in more detail. We found that NaHCO<sub>3</sub> and EtOH were the best base and solvent, respectively, affording *spiro*-aminal **8e** in 47% yield (entry 7). These conditions were also applicable to a gram-scale reaction of PMB urea **7f** to afford *spiro*-product **8f** in a similar yield (entry 8). Accordingly, we established a stereocontrolled synthesis of the BC ring of STX, including an unprecedented *spiro*-aminal structure, through a Br<sup>+</sup>-initiated cascade cyclization reaction by the judicious choice of substituent R.

Encouraged by the successful construction of the BC ring structure, we focused on the synthesis of the A ring from **8f** possessing a readily deprotectable PMB group (Scheme 3). As our preliminary experiments showed that the *gem*-CBr<sub>2</sub> group was unstable under several conditions, we initially investigated its transformation into a more stable hydroxy function. However, the attempted formal hydrolysis of *gem*-CBr<sub>2</sub> under previously reported radical-mediated conditions (Ac<sub>2</sub>O, Et<sub>3</sub>N, and air in CH<sub>2</sub>Cl<sub>2</sub>/H<sub>2</sub>O)<sup>12</sup> was unsuccessful, producing a mixture of unidentified products. After numerous experiments, we discovered that monoBoc-protected guanidine **10**, prepared by treatment of **8f** with periodic acid (1 equiv), undergoes a radical-mediated oxygenation under modified conditions (Ac<sub>2</sub>O, Et<sub>3</sub>N, and air in CH<sub>3</sub>CN/H<sub>2</sub>O) to provide a mixture of the corresponding ketone and enol acetate, both of which possess an acetate moiety on the guanidine fragment.<sup>20</sup> Reduction of the mixture with NaBH<sub>4</sub> yielded acetate **11** as the single product<sup>21</sup> in good overall yield. The protected diol in **11** was cleaved with periodic acid (2 equiv) to form cyclic urea **12** with an *N,O*-acetal as a single diastereomer. A nitrile was introduced using acetone cyanohydrin under Mitsunobu-type conditions (TMAD and Me<sub>3</sub>P),<sup>22</sup> furnishing single diastereo-

Table 1. Br<sup>+</sup>-Triggered Cascade Cyclization of 7b–f

entry	substrate		conditions			yield (%)	
	R		PyHBr <sub>3</sub> (equiv)	base	solvent(s)	8b–f	9b–f
1	7b:	C(=NBoc)NHBoc	1.3	K <sub>2</sub> CO <sub>3</sub>	CH <sub>2</sub> Cl <sub>2</sub> –H <sub>2</sub> O	8b 0	9b 66
2	7b:		3.0	K <sub>2</sub> CO <sub>3</sub>	CH <sub>2</sub> Cl <sub>2</sub> –H <sub>2</sub> O	8b 0	9b 62
3	7c:	C(=NTces)SMe	1.3	K <sub>2</sub> CO <sub>3</sub>	CH <sub>2</sub> Cl <sub>2</sub> –H <sub>2</sub> O	8c 0	9c 69
4	7c:		3.0	K <sub>2</sub> CO <sub>3</sub>	CH <sub>2</sub> Cl <sub>2</sub> –H <sub>2</sub> O	complex mixture	
5	7d:	C(=S)NHBn	1.3	K <sub>2</sub> CO <sub>3</sub>	CH <sub>2</sub> Cl <sub>2</sub> –H <sub>2</sub> O	complex mixture	
6	7e:	CONHBn	3.0	K <sub>2</sub> CO <sub>3</sub>	CH <sub>2</sub> Cl <sub>2</sub> –H <sub>2</sub> O	8e 23	9e 14
7	7e:		5.0	NaHCO <sub>3</sub>	EtOH	8e 47	9e trace
8	7f:	CONHPMB	5.0	NaHCO <sub>3</sub>	EtOH	8f 46	9f trace

Scheme 3. Synthesis of the A Ring and Total Synthesis of dc- $\alpha$ -STXol (2)

meric product **13**,<sup>23</sup> which contains all the carbons necessary for the synthesis of the STX skeleton. When the nitrile moiety in **13** was hydrolyzed by heating with hydrochloric acid, concomitant elimination of all protective groups was observed. The resulting carboxylic acid was methylated with hydrogen chloride in MeOH and **14a** was acetylated to provide tetraacetate **14b** in moderate overall yield from **13**. Finally, *O*-ethylisourea **15**, obtained in 53% yield from the reaction of **14b** with EtOTf,<sup>4c</sup> was treated with LiBH<sub>4</sub>.<sup>24</sup> An ammonolysis of the resulting *O*-ethylisourea furnished (+)-dc- $\alpha$ -STXol (**2**) in 65% overall yield from **15**. All spectroscopic data obtained were consistent with previously reported data.<sup>10</sup>

In summary, we have developed a short synthetic route for the formation of the STX skeleton, which relied on a newly designed cascade bromocyclization reaction of the internal

alkyne. This one-pot process provides an easily accessible route to a *spiro*-aminal structure containing a five-membered guanidine and pyrrolidine corresponding to the BC ring, which has not been prepared before. This new route is thus based on a strategy that is substantially different from previously reported synthetic approaches. Moreover, this study provides access to several STX analogues that differ with respect to the substituents at the C-6 position. This feature may be a key element for the development of subtype-selective inhibitors of VGSCs. In this study, we also have expanded the range of utility of the Br<sup>+</sup>-triggered cascade cyclization of alkyne with two heteroatom nucleophiles in the synthesis of heterocyclic compounds containing not only *N,O*-acetal<sup>25</sup> and *O,O*-acetal<sup>26</sup> but also the aminal structure. The synthesis of other STX analogues via this new route and further applications of this cascade cyclization reaction are currently ongoing in our laboratory.

## ■ ASSOCIATED CONTENT

### Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.orglett.6b03262.

Experimental procedures and spectral data (PDF)

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### Notes

The authors declare no competing financial interest.

## ■ ACKNOWLEDGMENTS

This work was supported by the Grant-in-Aid for Scientific Research on Innovative Areas “Chemical Biology of Natural Products”<sup>27</sup> from MEXT, and the Daiichi Sankyo Foundation of Life Science. S.U. is a research fellow of the Japan Society for the Promotion of Science (DC1).

## REFERENCES

- (1) (a) Schantz, E. J.; Ghazarossian, V. E.; Schnoes, H. K.; Strong, F. M.; Springer, J. P.; Pezzanite, J. O.; Clardy, J. *J. Am. Chem. Soc.* **1975**, *97*, 1238–1239. (b) Bordner, J.; Thiessen, W. E.; Bates, H. A.; Rapoport, H. *J. Am. Chem. Soc.* **1975**, *97*, 6008–6012.
- (2) (a) Salgado, V. L.; Yeh, J. Z.; Narahashi, T. *Ann. N. Y. Acad. Sci.* **1986**, *479*, 84–95. (b) Llewellyn, L. E. *Nat. Prod. Rep.* **2006**, *23*, 200–222.
- (3) Thottumkara, A. P.; Parsons, W. H.; Du Bois, J. *Angew. Chem., Int. Ed.* **2014**, *53*, 5760–5784.
- (4) (a) Fleming, J. J.; Du Bois, J. *J. Am. Chem. Soc.* **2006**, *128*, 3926–3927. (b) Fleming, J. J.; McReynolds, M. D.; Du Bois, J. *J. Am. Chem. Soc.* **2007**, *129*, 9964–9975. (c) Mulcahy, J. V.; Du Bois, J. *J. Am. Chem. Soc.* **2008**, *130*, 12630–12631. (d) Mulcahy, J. V.; Walker, J. R.; Merit, J. E.; Whitehead, A.; Du Bois, J. *J. Am. Chem. Soc.* **2016**, *138*, 5994–6001.
- (5) (a) Iwamoto, O.; Koshino, H.; Hashizume, D.; Nagasawa, K. *Angew. Chem., Int. Ed.* **2007**, *46*, 8625–8628. (b) Iwamoto, O.; Shinohara, R.; Nagasawa, K. *Chem. - Asian J.* **2009**, *4*, 277–285. (c) Iwamoto, O.; Nagasawa, K. *Org. Lett.* **2010**, *12*, 2150–2153. (d) Iwamoto, O.; Akimoto, T.; Nagasawa, K. *Pure Appl. Chem.* **2012**, *84*, 1445–1453. (e) Wang, C.; Oki, M.; Nishikawa, T.; Harada, D.; Yotsu-Yamashita, M.; Nagasawa, K. *Angew. Chem., Int. Ed.* **2016**, *55*, 11600–11603.
- (6) Bhonde, V. R.; Looper, R. E. *J. Am. Chem. Soc.* **2011**, *133*, 20172–20174.
- (7) (a) Tanino, H.; Nakata, T.; Kaneko, Y.; Kishi, Y. *J. Am. Chem. Soc.* **1977**, *99*, 2818–2819. (b) Hong, C. Y.; Kishi, Y. *J. Am. Chem. Soc.* **1992**, *114*, 7001–7006.
- (8) (a) Jacobi, P. A.; Martinelli, M. J.; Polanc, S. *J. Am. Chem. Soc.* **1984**, *106*, 5594–5598. (b) Martinelli, M. J.; Brownstein, A. D.; Jacobi, P. A.; Polanc, S. *Croat. Chem. Acta* **1986**, *59*, 267–295.
- (9) Sawayama, Y.; Nishikawa, T. *Angew. Chem., Int. Ed.* **2011**, *50*, 7176–7178.
- (10) Onodera, H.; Satake, M.; Oshima, Y.; Yasumoto, T.; Carmichael, W. W. *Nat. Toxins* **1997**, *5*, 146–151.
- (11) To the best of our knowledge, only a few synthetic methods leading to an arene-unfused 5/5 spiro-aminal system have been reported. (a) Büchel, K. H.; Bocz, A. K.; Korte, F. *Chem. Ber.* **1966**, *99*, 724–736. (b) Tsuge, O.; Watanabe, H.; Masuda, K.; Yousif, M. M. *J. Org. Chem.* **1979**, *44*, 4543–4547. (c) Kalaitzakis, D.; Antonatou, E.; Vassilikogiannakis, G. *Chem. Commun.* **2014**, *50*, 400–402. It is reported that 1,6-diazaspiro[4.4]nonane was unstable leading to the corresponding ring-opened iminoamine. (d) Denisenko, S. N.; Pasch, E.; Kaupp, G. *Angew. Chem., Int. Ed. Engl.* **1989**, *28*, 1381–1383 and references therein.
- (12) Kimura, R.; Sawayama, Y.; Nakazaki, A.; Miyamoto, K.; Uchiyama, M.; Nishikawa, T. *Chem. - Asian J.* **2015**, *10*, 1035–1041.
- (13) Yadav, J. S.; Chander, M. C.; Rao, C. S. *Tetrahedron Lett.* **1989**, *30*, 5455–5458.
- (14) Yamaguchi, M.; Hirao, I. *Tetrahedron Lett.* **1983**, *24*, 391–394.
- (15) For details, see the [Supporting Information](#).
- (16) (a) Sawayama, Y.; Nishikawa, T. *Synlett* **2011**, *2011*, 651–654. (b) Sawayama, Y.; Nishikawa, T. *Yuki Gosei Kagaku Kyokaiishi* **2012**, *70*, 1178–1186.
- (17) The structures of **9b** to **9e** were assigned on the basis of a 2D NMR spectroscopic analysis.
- (18) We reasoned that the Boc groups of the acyclic guanidine should shield the *N*-Boc enamine moiety of **9b** from Br<sup>+</sup>, thus promoting the formation of **9b**. Long-time exposure to an excess of the reagents resulted in the decomposition of **9b**.
- (19) CCDC-1496945 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via [www.ccdc.cam.ac.uk/data\\_request/cif](http://www.ccdc.cam.ac.uk/data_request/cif).
- (20) Conventional conditions using AgNO<sub>3</sub> in aqueous CH<sub>3</sub>CN did not afford the desired ketone, but an aminoimidazole as the major product.
- (21) The configuration of C-12 could not be assigned; an  $\alpha$  was tentatively assigned by comparison with the NMR spectra of **2**.
- (22) Tsunoda, T.; Uemoto, K.; Nagino, C.; Kawamura, M.; Kaku, H.; Ito, S. *Tetrahedron Lett.* **1999**, *40*, 7355–7358.
- (23) The stereochemistry at the C-6 position was determined on the basis of the coupling constant ( $J = 0$  Hz) between H6 and H5 in the <sup>1</sup>H NMR spectra, which is identical to that of the natural product ( $J = 0$  Hz).
- (24) Under these conditions, the methyl ester was reduced to the corresponding hydroxymethyl group and the acetyl groups were removed.
- (25) (a) Nakazaki, A.; Ishikawa, Y.; Sawayama, Y.; Yotsu-Yamashita, M.; Nishikawa, T. *Org. Biomol. Chem.* **2014**, *12*, 53–56. (b) Nakazaki, A.; Nakane, Y.; Ishikawa, Y.; Yotsu-Yamashita, M.; Nishikawa, T. *Org. Biomol. Chem.* **2016**, *14*, 5304–5309.
- (26) Nakazaki, A.; Nakane, Y.; Ishikawa, Y.; Nishikawa, T. *Heterocycles* **2015**, *91*, 1157–1167.
- (27) Ueda, M. *Chem. Lett.* **2012**, *41*, 658–666.