Tetrodotoxin Framework Construction from Linear Substrates Utilizing a $Hg(OTf)_2$-Catalyzed Cycloisomerization Reaction: Synthesis of the Unnatural Analogue 11-*nor*-6,7,8-Trideoxytetrodotoxin

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Tetrodotoxin Framework Construction from Linear Substrates Utilizing a $Hg(OTf)_2$-Catalyzed Cycloisomerization Reaction: Synthesis of Unnatural Analogue 11-*nor*-6,7,8-Trideoxytetrodotoxin

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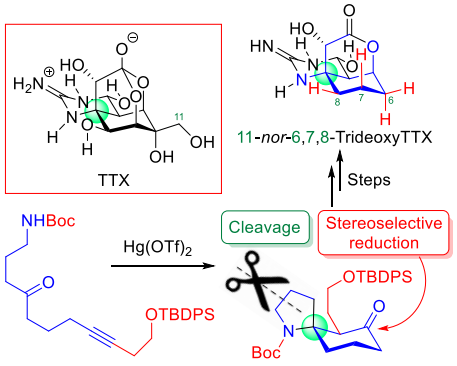
Supporting Information Placeholder

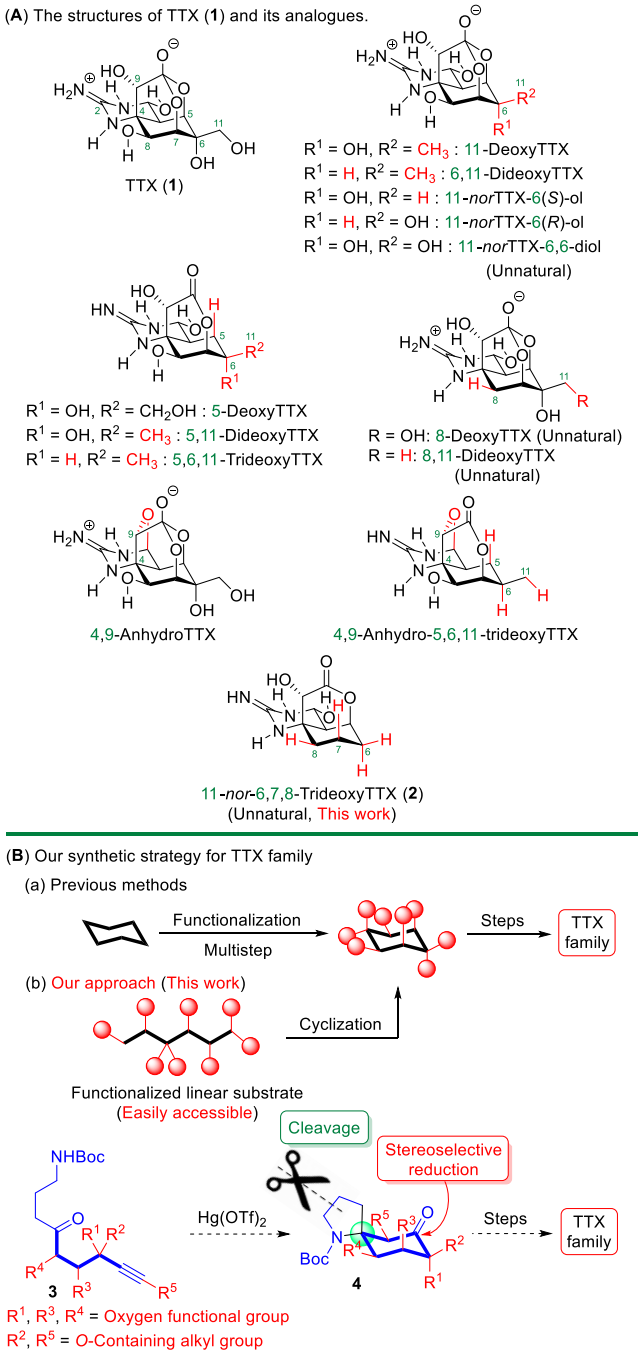


ABSTRACT: In this contribution, we propose a new synthetic approach to tetrodotoxin (TTX), one of the most famous marine toxins, that, after first preparing a functionalized linear substrate, forms the cyclohexane core from the substrate utilizing our mercuric triflate ($Hg(OTf)_2$)-catalyzed cycloisomerization reaction. The concept was applied to the synthesis of 11-*nor*-6,7,8-trideoxyTTX and 11-*nor*-4,9-anhydro-6,7,8-trideoxyTTX, unnatural TTX analogues, demonstrating the validity of our new approach.

Tetrodotoxin (**1**, TTX, Scheme 1A) is one of the best-known marine natural products and was first isolated from the ovaries of globefish by Tahara in 1909.[1] Its unique and complex structure was determined by three groups,[2] and was subsequently established via X-ray crystallographic analysis of its derivative.[3] The structural features that contain a dioxa-adamantane skeleton functionalized by a number of hydroxy groups and a cyclic guanidine in a small molecule make it the most difficult synthetic target in terms of total synthesis. TTX also exhibits a specific block of sodium ion influx through the sodium ion channel proteins.[4] The TTX family includes various analogues, largely deoxy and anhydro types (Scheme 1A).[5] The total synthesis of TTX and its natural analogues has been conducted several times,[6] whereas the synthesis of its unnatural analogues has also been reported in terms of the studies on the biochemical behavior of compounds with substituent patterns unavailable in natural form. For example, Isobe and Nishikawa et al. reported the synthesis of 8,11-dideoxyTTX[6l-n] and 8-deoxyTTX[6q] as unnatural analogues and demonstrated the role of the hydroxy group at the C8 position in sodium channel inhibition. Therefore, establishing a synthetic pathway for TTX analogues is crucial to elucidate the inhibition activity of voltage-gated sodium channels and the biosynthetic pathways for TTX.

In fact, the total synthesis of TTX has been reported by five research groups.[6] All of the synthetic strategies reported thus far have adopted the construction of the cyclohexane core at the early stage; however, the transformations of dense functional groups on the cyclohexane core resulted in numerous reaction steps due to the attendant steric hindrance (Scheme 1B). Considering this, we aimed to synthesize the TTX-type functionalized linear substrate at the early stage of synthesis to minimize the number of extra steps of transformation on the congested cyclohexane core. Following this, the mid-stage cyclization could construct the cyclohexane core and the latter conversion could facilitate the access to TTX. The key cyclization is our $Hg(OTf)_2$-catalyzed cycloisomerization reaction[7] that constructs a 1-azaspiro[4.5]decane skeleton with a nitrogen-containing tetrasubstituted carbon from linear substrates in high stereoselectivity and good yield via a Petasis–Ferrier-type reaction. In this plan, the

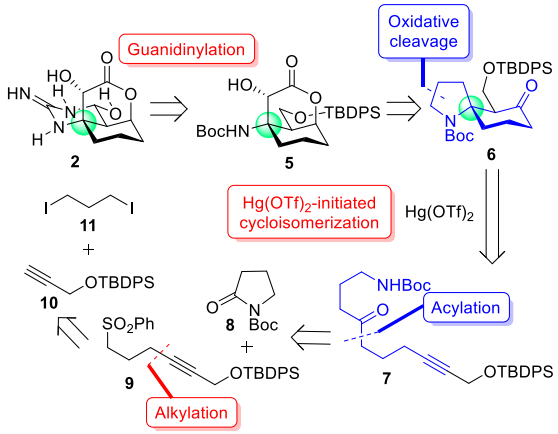
Scheme 1. Structures of TTX (1) and Its Analogues, and (B) Our Synthetic Strategy for the TTX Family



functionalized linear substrate **3** should be treated with $Hg(OTf)_2$ to yield the key intermediate spirocycle **4**, which would be led to the TTX family through the stereoselective reduction of a carbonyl moiety and the carbon–carbon bond oxidative cleavage on a pyrrolidine ring followed by intramolecular cyclization to construct a lactone ring. If certain linear substrates with various oxygen functional groups could be synthesized, the synthetic strategy for the desired TTX analogues could be established by the cycloisomerization producing the corresponding spirocycles. In this article, we report the synthesis of 11-*nor*-6,7,8-trideoxyTTX (**2**), an unnatural analogue of TTX, as the first model compound (Scheme 1A). Since the 7-deoxy analogue has been produced neither naturally nor through synthesis, the role of the hydroxy group at the C7 position in sodium channel inhibition remains unclear. The validity of our synthetic strategy would be confirmed by the synthesis of analoue **2**.

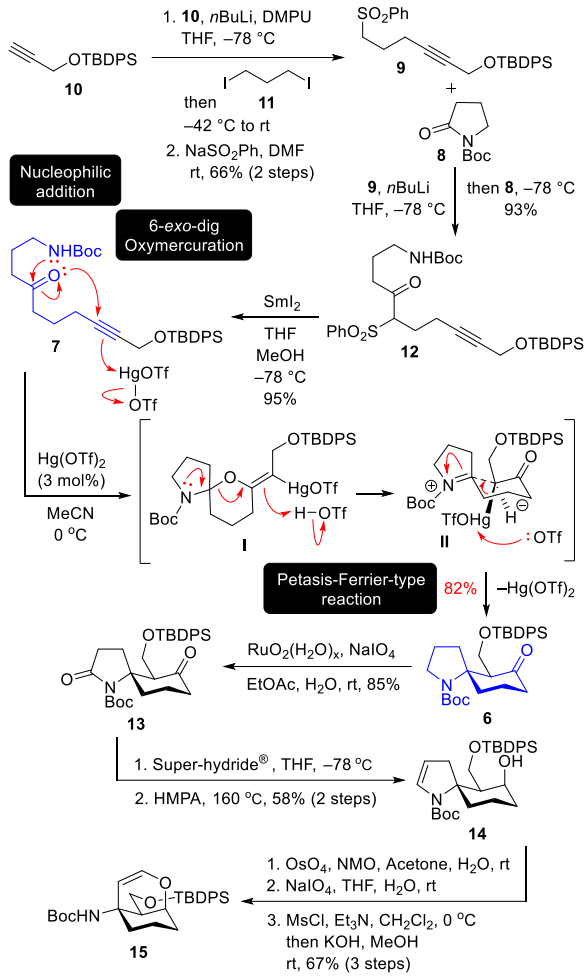
The retrosynthetic analysis of analogue **2** is shown in Scheme 2. Here target **2** would be synthesized through guanidinylation from lactone **5**, which would be derived from spirocycle **6** via the carbon–carbon bond oxidative cleavage. Compound **6** with a nitrogen-containing tetrasubstituted carbon would be then constructed by the key cycloisomerization of linear ynone **7**, which would be prepared through acylation of sulfone **9** with easily accessible pyrrolidinone **8**. We predicted that compound **9** would be synthesized by alkylation of a lithium acetylide of the known alkyne **10** with 1,3-diiodopropane (**11**).

Scheme 2. Retrosynthetic Analysis of 11-*nor*-6,7,8-TrideoxyTTX (2)



The synthesis commenced with the known alkyne **10**, derived from commercially available 2-propyn-1-ol (Scheme 3).[8] Sulfone **9** was synthesized through alkylation of a lithium acetylide of **10** with commercially available 1,3-diiodopropane (**11**) and subsequent sulfonylation. Following the acylation of an α-anion of **9** with the known pyrrolidinone **8**,[9] which can be easily prepared from commercially available 2-pyrrolidone, the cyclization precursor **7** was prepared through SmI_2-mediated desulfonylation[10] of **12**. The key $Hg(OTf)_2$-catalyzed cycloisomerization reaction[7] of **7** produced the desired spirocycle **6** in a stereoselective manner (82%).[11,12] The aminoketal **I** would be formed through a 6-*exo*-dig intramolecular oxymercuration to the alkyne π-electron activated by coordination of the catalyst followed by the nucleophilic addition of the nitrogen function. The intermediate **I** could be cleaved via protonation with the generated TfOH to give iminium ion intermediate **II** (Petasis–Ferrier-type cyclization). The construction of a carbocycle would provide spirocycle **6** with regeneration of the catalyst. Considering certain chair-like transition states, the desired **6** would diastereoselectively be obtained by way of the more stable transition state **II** with less steric repulsion.[13] Following the oxidation using RuO_4 to afford amide **13**,[14] the hydride reduction using Super-hydride® produced the desired diol, which was heated in HMPA to give enecarbamate **14** as a sole diastereomer.[15] Next, we

Scheme 3. Synthesis of Enol Ether 15

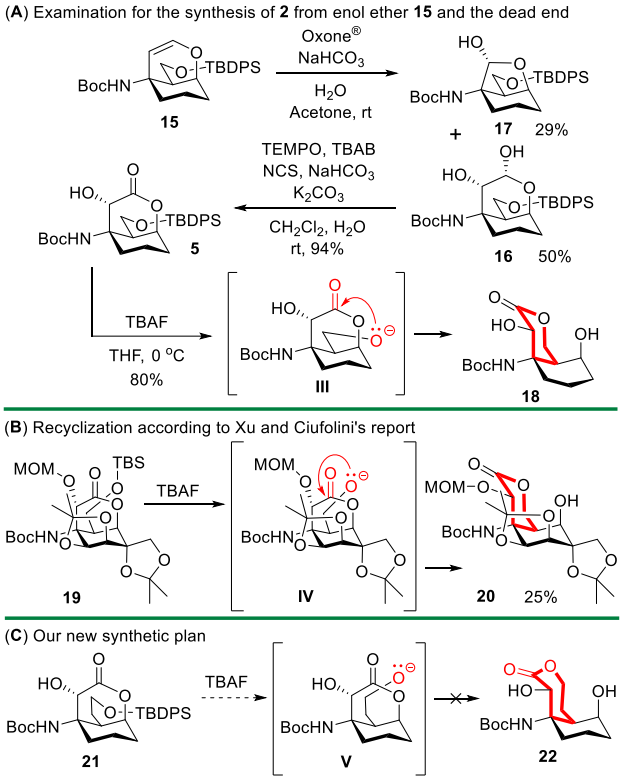


intended to cleave a double bond in **14**. After **14** was treated with OsO_4 and NMO, the oxidative cleavage of the resultant diol using $NaIO_4$ followed by intramolecular cyclization resulted in the desired lactol. Following dehydration using MsCl and Et_3N, the desired enol ether **15** was obtained via removal of a formyl group.

Next, we focused on the synthesis of analogue **2** from **15** (Scheme 4A). Dihydroxylation using Oxone® and $NaHCO_3$ produced the desired diol **16** in 50% yield, along with byproduct **17** which would oxidatively be cleaved from **16** (29%).[16,17] The selective TEMPO oxidation[18] of **16** produced lactone **5** in good yield (94%). However, when a *tert*-butyldiphenylsilyl (TBDPS) protecting group in **5** was removed using tetra-*n*-butylammonium fluoride (TBAF), only undesired lactone **18** was obtained in 80% yield due to the unexpected intramolecular nucleophilic attack. In 2015, Xu and Ciufolini reported a similar recyclization reaction in a formal synthesis of TTX (Scheme 4B).[19] To prevent this unwanted nucleophilic attack, we increased the number of methylene units in the side chain when synthesizing lactone **21** (Scheme 4C). It is presumed that the recyclization wouldnot proceed by the TBDPS deprotection in **21** because the seven-membered lactone **22** is difficult to generate.

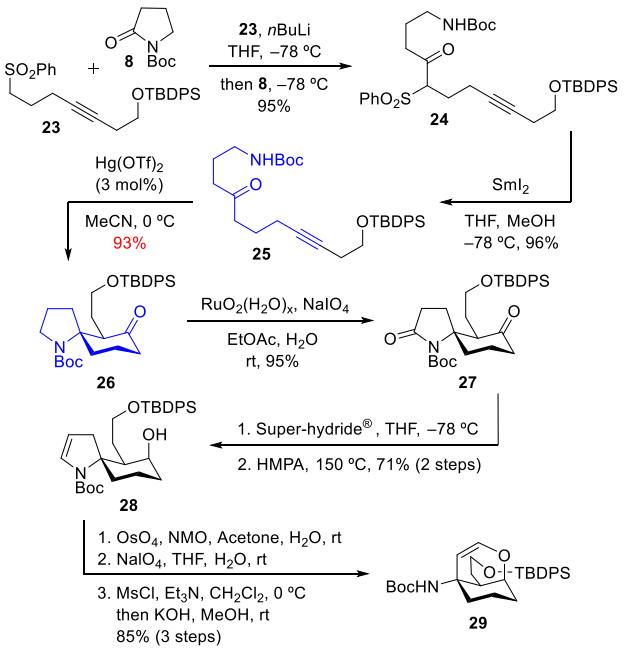
Enol ether **29,** which increased a methylene unit in the side chain, was synthesized from the known sulfone **23**[7c] and pyrrolidinone **8** (Scheme 5). Compound **29** was

Scheme 4. (A) Synthetic Study on Producing Target Analogue 2 from Enol Ether 15, (B) Recyclization According to Xu and Ciufolini's Report, and (C) Our New Synthetic Plan



prepared according to the same pathway as shown in Scheme 3. As the key reaction, the cycloisomerization reaction[7] of ynone **25** yielded the desired spirocycle **26** in high yield (93%) and in a diastereoselective manner.

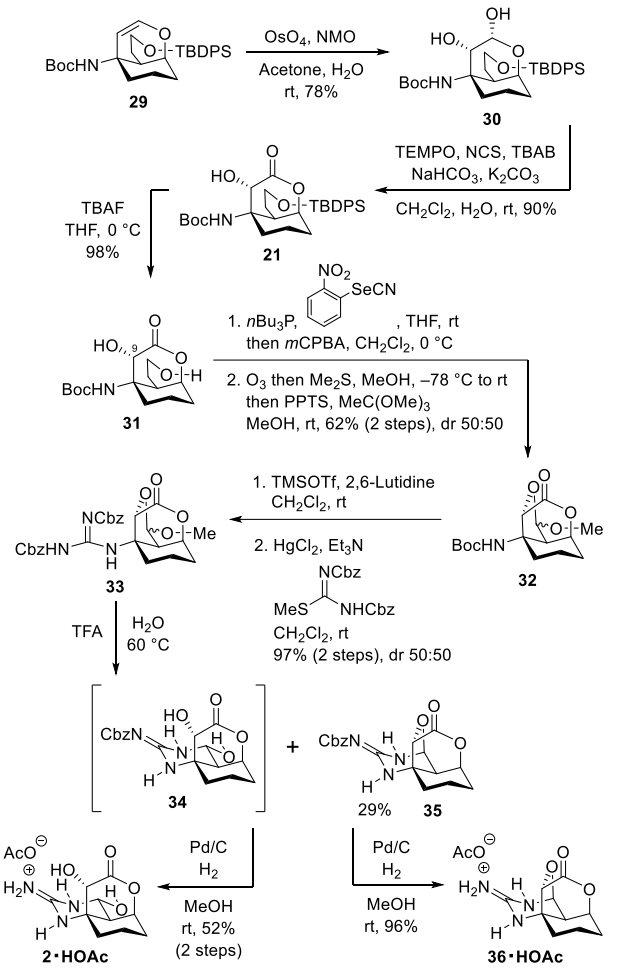
Scheme 5. Synthesis of Enol Ether 29.



We tried to synthesize analogue **2** from enol ether **29** as shown in Scheme 6. In the next step, dihydroxylation of **29** with OsO_4 produced diol **30** as a single diastereomer[20] before the selective TEMPO oxidation[18] of **30** provided lactone **21** (90%), along with diketone as a minor byproduct (9%). As expected, the desired alcohol **31** was obtained in high yield (98%) through the deprotection of a TBDPS group utilizing TBAF at 0 °C,[21] while the unwanted seven-membered lactone **22** was not detected. Compound **31** was transformed to acetal **32** via the following sequence: 1) Nishizawa–Grieco elimination[22] to construct a vinylic group, 2) ozonolysis of a vinylic group, and 3) acetal formation using PPTS and $MeC(OMe)_3$. Following removal of a Boc group with TMSOTf and 2,6-lutidine,[6o] guanidinylation of the resultant amine produced **33** as a mixture of diastereromers in high yield (97%). Proceeding with reference to Fukuyama's report,[6j] the deprotection under acidic conditions led to a mixture of mono-Cbz-protected precursor **34** and its anhydro form **35**. Finally, we managed to synthesize our target analogue, 11-*nor*-6,7,8-trideoxyTTX acetate (**2·HOAc**), through hydrogenolysis of a Cbz group and HPLC purification.[5n] An anhydro form of **2·HOAc**, 11-*nor*-4,9-anhydro-6,7,8-trideoxyTTX acetate (**36·HOAc**), was also obtained from **35** via the same process.

In summary, we have achieved the syntheses of 11-*nor*-6,7,8-trideoxyTTX, an unnatural tetrodotoxin analogue, and its anhydro form, 11-*nor*-4,9-anhydro-6,7,8-trideoxyTTX,[23] from a spirocyclic compound constructed through our $Hg(OTf)_2$-catalyzed cycloisomerization reaction of a linear substrate. The success of these syntheses encourages our new approach to TTX shown in Scheme 1B. The application of our synthetic strategy to natural TTX and other deoxy analogues is currently underway in our laboratory.

Scheme 6. Synthesis of 11-*nor*-6,7,8-TrideoxyTTX Acetate (2·HOAc) and Anhydro Form 36·HOAc



ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI. All experimental procedures, the spectroscopic data, and copies of ^1H- and ^{13}C-NMR spectra.

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Notes

The authors declare no competing financial interest.

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(20) Dihydroxylation of **29** using Oxone® and $NaHCO_3$ gave the desired **30** in 64% yield, along with lactol which would oxidatively be cleaved from **30** (25%).

(21) The TBDPS deprotection at room temperature afforded a mixture of two diastereomers, diol **31** and its 9-epimer (dr 59:41), see the SI. We selectively synthesized 9-epimer (dr 90:10) through the $NaBH_4$ reduction of diketone which produced as a byproduct in the oxidation of **30**, followed by the deprotection of a TBDPS protecting group, to obtain the spectral data of 9-epimer. The stereochemistry of 9-epimer was clearly assigned based on the NOESY spectrum; see the SI.

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