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A Synthesis of (+)-Saxitoxin

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The unassuming molecular dimensions of the poison most notably associated with oceanic red tides, (+)-saxitoxin (STX), belie its complexity as a target for chemical synthesis and its acute toxicity as a paralytic agent (Figure 1).[1] The structure, a tricyclic skeleton uniquely adorned with nitrogen and oxygen heteroatoms, has contested those interested in its de novo assembly as well as its isolation and characterization.[2,3] Nevertheless, two formidable efforts describe salient paths for preparing this compound.[4,5] Our own interest in STX followed for two reasons: (1) as a vehicle by which to advance new strategies for crafting polar, nitrogen-rich functional groups in the most demanding circumstances; (2) as a blueprint for the design of pharmacological tools that could help elucidate pathways for electrical conduction in excitable cells. The lethality of STX derives from its ability to block selectively cation influx through voltage-dependent Na^+ ion channels.[6] Although its action has been known and exploited for some time, the molecular details of STX lodged in the channel pore remain ambiguous and subject to debate.[7] Such knowledge, however, could empower the development of new small molecules for the manipulation and study of ion channel function. As such, a synthesis of STX was undertaken with the goal of forwarding chemical methodologies in a preparative route that offered sufficient flexibility for modifying the toxin's design.

An initial examination of STX reveals that both C4 and C12 centers reside formally at the ketone oxidation level (Figure 1). At physiological pH, the equilibrium between the C12-ketone and its hydrated form strongly favors the latter.[8] Retrosynthetic disconnection of the adjacent spiro-aminal junction posits a cyclodehydration reaction in which both guanidines are condensed onto a ketone at C4.[9] In principle, the stereochemical configuration of the attendant C5 and C6 centers would predispose formation of the requisite C4 epimer. To further enable this plan, one of the two guanidines would be incorporated within a nine-membered ring **1**. Although almost no precedent for the assembly of medium-sized guanidine rings is available, we anticipated that pseudothiourea **2** could be condensed with a 1° amine at C6, possibly through the intermediacy of a reactive carbodiimide. The overarching strategy to assemble STX thus reduces a complicated problem in cyclic stereocontrolled synthesis to the seemingly more manageable task of constructing an acyclic core **2**.

The assembly of stereodefined, polyfunctionalized amines as exemplified by **2** provides a unique opportunity to challenge our current methods for catalytic C−H amination. In wanting to access such complex intermediates, we have devised a strategy that capitalizes on a novel class of *N*,*O*-acetal heterocycles made readily available through Rh-catalyzed sulfamate ester C−H insertion.[10] The particular *N*,*O*-acetal **3** needed for the STX synthesis is easily prepared on multigram scale from commercial (*R*)-glycerol acetonide.[11] Lewis acid-promoted addition of a zinc-acetylide to **3** afforded oxathiazinane dioxide **4** (R = CH_2OTs) as a single diastereomer. The advantage of this heterocycle is both as a masking group of the basic amine and as a latent electrophile, which may

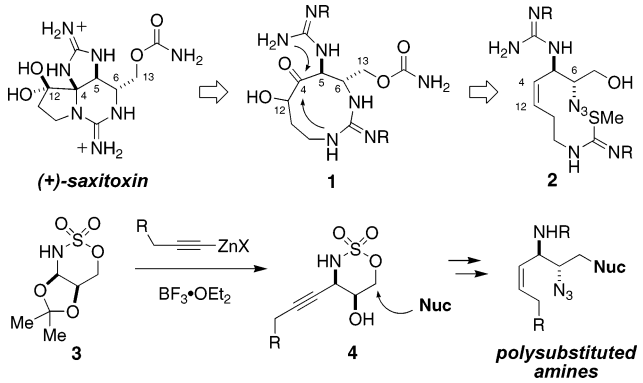

Figure 1. Deconstructing (+)-saxitoxin to a problem of acyclic stereocontrolled synthesis: the oxathiazinane strategy.

be activated for ring opening at an appropriate stage. Accordingly, the *N*,*O*-acetal strategy offers a general approach to multiply substituted amine derivatives that transcends its usefulness for the construction of STX.

The preparation of the desired nine-membered ring guanidine **1** follows from oxathiazinane **4** through a series of straightforward functional group transformations (Scheme 1). Notable steps in this sequence include the assembly of pseudothiourea **6** using MeS(Cl)C═NMbs, a reagent specifically formulated for this work.[12] In addition, sequential reaction of oxathiazinane **6** with Cl_2C═NMbs and $(Me_3Si)_2NH$ installs the first of two guanidines while serving to activate the heterocyclic ring for subsequent hydrolytic opening.[13,14] Collectively, the conversion of **4** to **8** covers 11 steps and provides a stereodefined acyclic intermediate bearing all of the required components for assembling the tricyclic framework of STX.

With very little precedent guiding our efforts to form the medium-sized ring guanidine, a number of reaction conditions were explored.[15] The most successful of those tested afforded a 65% yield (two steps) of the needed product **9**. In the devised protocol, reduction of azide **8** with Me_3P was followed by immediate exposure of this compound to $AgNO_3/Et_3N$.[12] The latter conditions presumably trigger formation of a reactive *N*-sulfonylcarbodiimide, which in turn is intercepted by the pendant C6-amine. In the product heterocycle **9**, the highly polar nature of the two guanidine moieties is nicely masked by Mbs protection; functionalization of the C13-alcohol in **9** as the obligatory 1° carbamate is therefore easily conducted ($Cl_3CC(O)NCO$).[16]

Difficulties encountered in the synthesis of **9** would prove, by comparison, less perilous than those confronted in the oxidation of **10**. The availability of select methods for effecting four-electron alkene ketohydroxylation led us to consider such a transformation;[17] tautomeric control of the α-ketol isomer, however, loomed as a conspicuous uncertainty in this plan. In fact, reactions performed with the known combination of OsO_4 and ^{t}BuOOH gave only the undesired bicyclic structure **13** (Figure 2).[17c] Fortunately, continued

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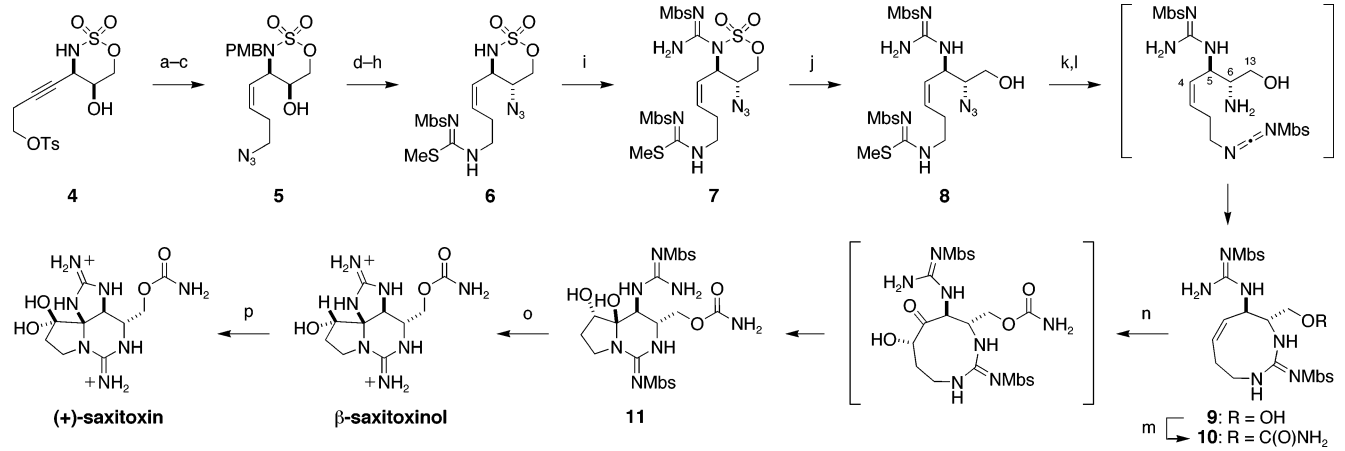
Scheme 1 [a]



[a] Conditions: (a) H_2, Pd/$CaCO_3$/Pb, THF; (b) NaN_3, $^nBu_4NI$, DMF, 90% (2 steps); (c) *p*-$MeOC_6H_4CH_2Cl$, $^nBu_4NI$, K_2CO_3, CH_3CN, 85%; (d) Me_3P, THF/H_2O; (e) MeS(Cl)C=NMbs, $^iPr_2NEt$, CH_3CN, 72% (2 steps); (f) Tf_2O, C_5H_5N, DMAP, CH_2Cl_2; (g) NaN_3, DMF, −15 °C, 70% (2 steps); (h) $(NH_4)_2Ce(NO_3)_6$, ^{t}BuOH/CH_2Cl_2, 74%; (i) KO^{t}Bu, Cl_2C=NMbs; then $(Me_3Si)_2NH$, 70% (+20% of **6**); (j) aq. CH_3CN, 70 °C, 95%; (k) Me_3P, THF/H_2O; (l) $AgNO_3$, Et_3N, CH_3CN, 65% (2 steps); (m) $Cl_3CC(O)NCO$, THF/CH_3CN, −78 °C; then K_2CO_3, MeOH, 82%; (n) 10 mol % of $OsCl_3$, Oxone, Na_2CO_3, EtOAc/CH_3CN/H_2O, 57%; (o) $B(O_2CCF_3)_3$, CF_3CO_2H, 82%; (p) DCC, C_5H_5N·HO_2CCF_3, DMSO, 70%. Mbs = *p*-$MeOC_6H_4SO_2$.

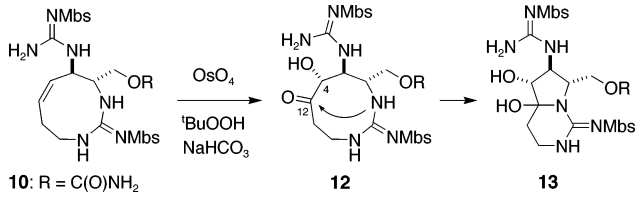


Figure 2. The regioisomeric α-ketol **12** gives an undesired result.

efforts to reverse this selectivity proved successful, as the use of catalytic $OsCl_3$ (10 mol %), Oxone, and Na_2CO_3 gave rise to 57% yield of the desired product **11** as a single stereoisomer. Rather remarkably, <5% of **13** is generated under these conditions. The interconversion of the two isomeric products when treated with acid does not occur, and thus it appears that selectivity in this reaction is kinetic in origin. While a cogent explanation for these contrasting discoveries cannot be offered at this time, the end result advantageously positions the proposed route for completion.

To deliver the STX core from **11** in a manner most efficient, conditions were sought to effect simultaneous cyclization of the five-membered ring guanidine and Mbs deprotection. Such was the case when **11** was treated with $B(O_2CCF_3)_3$, generating the known product, β-saxitoxinol, in 82% yield.[18] A two-step sequence beginning with the nine-membered ring **10** and ending with the fully assembled tricyclic frame of STX thus gives form to our initial retrosynthetic proposal. Finally, exposure of β-saxitoxinol to oxidative conditions previously described afforded the desired guanidinium poison.[19] Reverse-phase HPLC, with perfluorobutyric acid added to the eluent, gave pure samples of the synthetic material as the bis-$C_3F_7CO_2^-$ salt.[20] This material matched the physical data of natural (+)-STX in all respects. In addition, electrophysiology measurements against $Na_v1.4$ channels overexpressed in CHO cells furnished an IC_{50} value of 5 nM, consistent with literature reports.[1b,7c,21] The completed work, while highlighting several methodological inventions, now serves as an entry point for the construction of unique small molecules able to modify Na^+ channel function.

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Note Added after ASAP Publication. After this paper published ASAP, structure 1 in Figure 1 was corrected. The corrected version was published ASAP on March 3, 2006.

Supporting Information Available: Analytical data for all new compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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