Studies toward a Marine Toxin Immunogen: Enantioselective Synthesis of the Spirocyclic Imine of (−)-Gymnodimine

ARTICLE *in* ORGANIC LETTERS · DECEMBER 2005

Impact Factor: 6.36 · DOI: 10.1021/ol051840r · Source: PubMed

3 AUTHORS, INCLUDING:

Ziad Moussa
Taibah University
37 PUBLICATIONS **643** CITATIONS

SEE PROFILE

Daniel Romo
Baylor University
169 PUBLICATIONS **3,811** CITATIONS

SEE PROFILE

ORGANIC LETTERS

2005
Vol. 7, No. 23
5127−5130

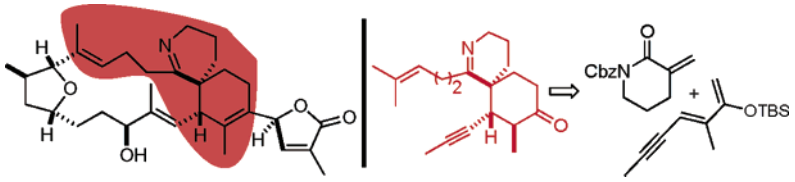
Studies toward a Marine Toxin Immunogen: Enantioselective Synthesis of the Spirocyclic Imine of (−)-Gymnodimine

Ke Kong, Ziad Moussa, and Daniel Romo*

Department of Chemistry, Texas A&M University, P.O. Box 30012, College Station, Texas 77842-3012
romo@mail.chem.tamu.edu

Received August 1, 2005 *(Revised Manuscript Received September 29, 2005)*

ABSTRACT



An enantioselective Diels–Alder reaction catalyzed by an Evans' copper–bis(oxazoline) complex was utilized to construct a highly functionalized spirolactam, a key intermediate in our projected total synthesis of the marine toxin, gymnodimine. Additional transformations, including a mild *N*-tosyl group deprotection, afforded a keto spirocyclic imine moiety, the proposed pharmacophore of gymnodimine. Thus, the prepared ketone is a potentially useful intermediate for conjugation to provide an immunogen for eventual monitoring of gymnodimine and congeners.

In 1994, oysters from Foveaux Strait, South Island, New Zealand, were found to be toxic and led to the isolation and structural elucidation of a novel marine toxin, (−)-gymnodimine (**1**) (Figure 1).[1] Subsequently, Blunt, Munro, and co-workers established the absolute stereochemistry of gymnodimine by X-ray crystallographic analysis of its *p*-bromobenzamide derivative.[2] More recently, two new analogues, gymnodimine B and C, have been reported.[3]

Gymnodimine and analogues exhibit unusual structural features, including a spirocyclic imine ring system and a trisubstituted tetrahydrofuran embedded within a 16-membered macrocycle. The presence of the spirocyclic imine places this marine toxin in the same family with the spirolides,[4] the pinnatoxins,[5] and prorocentrolide.[6] The toxicity of the spirolides has been attributed to the imine functionality as imine reduction leads to nontoxic derivatives.[7] However, the exact mode of action of these toxins

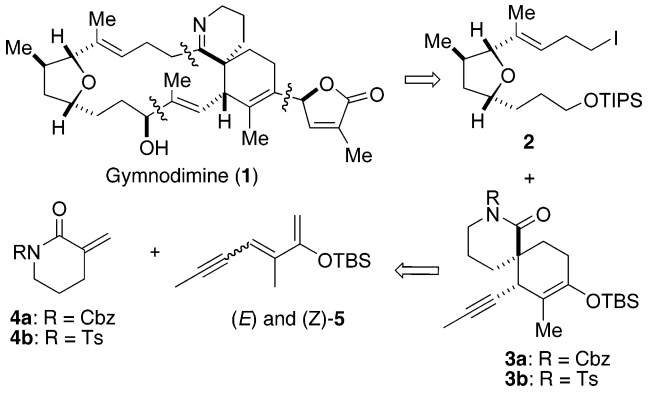


Figure 1. Retrosynthetic analysis of gymnodimine.

(1) Seki, T.; Masayuki, S.; Mackenzie, L.; Kaspar, H. F.; Yasumoto, T. *Tetrahedron Lett*. **1995**, *36*, 7093.
(2) Stewart, M.; Blunt, J. W.; Munro, M. H. G.; Robinson, W. T.; Hannah, D. J. *Tetrahedron Lett*. **1997**, *38*, 4889.
(3) (a) Miles, C. O.; Wilkins, A. L.; Stirling, D. J.; MacKenzie, A. L. *J. Agric. Food Chem.* **2000**, *48*, 1373. (b) Miles, C. O.; Wilkins, A. L.; Stirling, D. J.; MacKenzie, A. L. *J. Agric. Food Chem.* **2003**, *51*, 4838.
(4) Hu, T.; Curtis, J. M.; Oshima, Y.; Quilliam, M. A.; Walter, J. A.; Wright, W. M.; Wright, J. L. C. *J. Chem. Soc., Chem. Commun*. **1995**, 2159.
(5) Uemura, D.; Chou, T.; Haino, T.; Nagatsu, A.; Fukuzawa, S.; Zheng, S.; Chem, H. *J. Am. Chem. Soc*. **1995**, *117*, 1155.
(6) Torigoe, K.; Murata, M.; Yasumoto, T.; Iwashita, T. *J. Am. Chem. Soc*. **1988**, *110*, 7876.

10.1021/ol051840r CCC: $30.25
Published on Web 10/14/2005

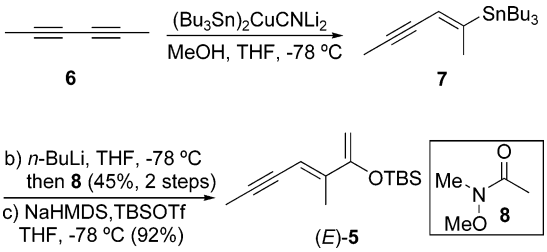
has not been elucidated as the only activity reported for the spirolides is weak activation of type L calcium channels (at 1.7 mM), and this activity does not correlate with the potent toxicity of these compounds. The potential for human consumption of shellfish contaminated with toxic concentrations of gymnodimine is prompting efforts to develop assays to monitor gymnodimine and congeners.[8] This is even more pressing given the recent finding that gymnodimine persists in shellfish for extended periods.[9]

In addition to bioactivity, the intriguing structures of gymnodimine (**1**, Figure 1) and related natural products have led to considerable attention from synthetic groups. An elegant total synthesis of pinnatoxin reported by Kishi employed an intramolecular Diels−Alder reaction to install the quaternary carbon of the spirocyclic core structure.[10] A recent extension of this strategy by the same group employing iminium dienophiles toward gymnodimine was recently disclosed.[11] Our initial studies demonstrated the potential of an intermolecular Diels−Alder process between an α-methylene lactam **4b** and dienes **5** to rapidly assemble the spirocyclic moiety common to the gymnodimines (Figure 1). This [4 + 2] cycloaddition was promoted by Et_2AlCl and proceeded in good yield and excellent diastereoselectivity.[12a] Subsequently, Murai[13] and White[14] disclosed their routes to the spirocyclic center involving Diels−Alder variants. Murai's intermolecular, double-diasteroselective Diels−Alder process employed Ellman's copper bis(sulfinyl)−imidoamidine complex[15] on an advanced chiral intermediate and proceeded with high yield and diastereoselectivity.[13b] In White's approach, an intermolecular Diels−Alder reaction employing an optically active diene was utilized to form the spirocyclic system, albeit with low diastereoselectivity.[14b] Brimble has also described approaches to the spirocyclic imine of gymnodimine.[16] Herein, we report the asymmetric synthesis of the spirocyclic moiety via an enantioselective Diels−Alder process and subsequent mild conversion to a model spirocyclic imine moiety of gymnodimine. This compound will be studied as an immunogen for the development of an assay to monitor the concentration of gymnodimine and congeners in shellfish and coastal regions.

Our retrosynthesis relies on butenolide annulation onto the spirocyclic intermediate **3**, coupling of tetrahydrofuran **2** with the spirocyclic imine precursor **3**, and finally macrocyclization to provide the core framework of gymnodimine (Figure 1). Building on our success in the racemic series, we envisioned an asymmetric version of the Diels−Alder reaction between α-methylene lactam **4** and diene **5** to provide optically active spirocyclic lactam **3**.[12a] There are relatively few examples of catalytic, asymmetric Diels−Alder processes employed in natural product total syntheses.[17]

Initial attempts to promote an enantioselective Diels−Alder reaction with the previously employed diene (*Z*)-**5** revealed a complete lack of reactivity with several chiral catalysts. We reasoned that the lack of reactivity may be due to the olefin geometry,[18] and thus pursued the synthesis of diene (*E*)-**5**. Synthesis of this diene commenced with stannylcupration[19] of 2,4-hexadiyne **6**, affording (*E*)-vinyl stannane **7** as the major isomer (89%, 5:2 mixture of regioisomers, Scheme 1). While radical and metal-catalyzed

Scheme 1. Synthesis of Diene (*E*)-**5**



hydrostannylations gave higher yields, the regioselectivity was less satisfactory.[20] Pd-catalyzed hydrostannylation gave the undesired regioisomer as the sole product. Tin−lithium exchange of stannane **7** followed by reaction with amide **8** gave the corresponding ketone. Subsequent enolization and silylation gave diene (*E*)-**5**.

After surveying several chiral catalysts, we found that Evans' copper−bis(oxazoline) hexafluoroantimonate complex (**9**)[21] promoted the Diels−Alder reaction of lactam **4a**

(7) Hu, T.; Curtis, J. M.; Walter, J. A.; Wright, J. L. C. *Tetrahedron Lett.* **1996**, *37*, 7671−7674.

(8) (a) McNabb, P.; Selwood A. I.; Holland P. T. *J. AOAC Int.* **2005**, 88, 761. (b) MacKenzie, L.; Holland, P.; McNabb, P. *Toxicon* **2002**, *40*, 1321.

(9) Stirling, D. *NZ J. Mar. Freshwat. Res.* **2001**, *35*, 851.

(10) McCauley, J. A.; Nagasawa, K.; Lander, P. A.; Mischke, S. G.; Semones, M. A.; Kishi, Y. *J. Am. Chem. Soc.* **1998**, *120*, 7647.

(11) Johannes, J. W.; Wenglowsky, S.; Kishi, Y. *Org. Lett.* **2005**, *7*, 3997.

(12) (a) Yang, J.; Cohn, S. T. Romo, D. *Org. Lett.* **2000**, *2*, 763. (b) Ahn, Y.; Cardenas, G. I.; Yang, J.; Romo, D. *Org. Lett.* **2001**, *3*, 751.

(13) (a) Ishihara, J.; Miyakawa, J.; Tsujimoto, T.; Murai, A. *Synlett* **1997**, 1417. (b) Tsujimoto, T.; Ishihara, J.; Horie, M.; Murai, A. *Synlett* **2002**, 399.

(14) (a)White, J. D.; Wang, G.; Quaranta, L. *Org. Lett.* **2003**, *5*, 4109. (b) White, J. D.; Wang, G.; Quaranta, L. *Org. Lett.* **2003**, *5*, 4983.

(15) Owens, T. D.; Hollander, F. J.; Oliver, A. G.; Ellman, J. A. *J. Am. Chem. Soc.* **2001**, *123*, 1539.

(16) Brimble, M. A.; Trzoss, M. *Tetrahedron* **2004**, *60*, 5613.

(17) (a) For a review of catalytic, asymmetric Diels−Alder reactions, see: Dias, L. C. *J. Braz. Chem. Soc.* **1997**, *8*, 289. (b) For more recent examples, see: Usuda, H.; Kuramochi, A.; Kanai, M.; Shibasaki, M. *Org. Lett.* **2004**, *6*, 4387 and references cited therein.

(18) In general, (*Z*)-dienes are known to be poor reactants in Diels−Alder reactions. However, some notable exceptions have been reported. For example, see: Roush, W. J.; Barda, D. A. *J. Am. Chem. Soc.* **1997**, *119*, 7402.

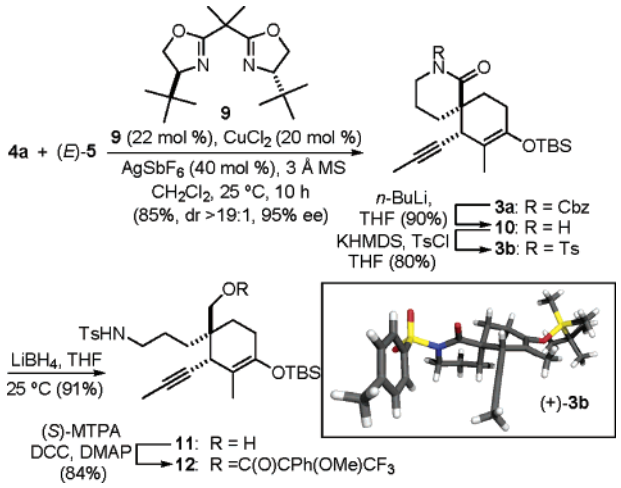
(19) Betzer, J.-F.; Ardisson, J., Lallemand, J.-Y.; Pancrazi, A. *Tetrahedron Lett.* **1997**, *38*, 13, 2279.

(20) For a review: Smith, N. D.; Mancuso, J.; Lautens, M. *Chem. Rev.* **2000**, *100*, 3257.

(21) (a) Evans, D. A.; Miller, S. J.; Lectka, T.; Matt, P. V. *J. Am. Chem. Soc.* **1999**, *121*, 7559. (b) Evans, D. A.; Barnes, D. M.; Johnson, J. S.; Lectka, T.; Matt, P. V.; Miller, S. J.; Murry, J. A.; Norcross, R. D.; Shaughnessy, E. A.; Campos, K. R. *J. Am. Chem. Soc.* **1999**, *121*, 7582. (c) For use of the triflate salt in related Diels−Alder reactions towards pinnatoxins, see: Ishihara, J.; Horie, M.; Shimada, Y.; Tojo, S.; Murai, A. *Synlett* **2002**, 404. (d) For related studies which appeared when our work was in progress, see: Brimble, M. A.; Crimmins, D.; Trzoss, M. *ARKIVOC* **2005**, *1*, 39.

and diene (*E*)-**5** to afford spirolactam **3a** as a single diastereomer in 85% yield (Scheme 2). The enantioselectivity

Scheme 2. Enantioselective Diels−Alder Reaction Leading to Spirolactam **3A**



of the process was determined initially by ^{1}H NMR using chiral shift reagents[22] and then independently verified by Mosher ester analysis of a derived alcohol. Attempted reduction of the Cbz-lactam led to partial deprotection of the Cbz group demonstrating the congested nature of the carbonyl carbon due to the α-quaternary center. In related studies, we found that addition of *n*-BuLi led to exclusive Cbz deprotection (vide infra, Scheme 3). This allowed a chemoselective deprotection and conversion to the *N*-tosyl lactam. Reduction to alcohol **11** with $LiBH_4$ proceed smoothly and this was converted to Mosher ester **12**. Analysis of the crude mixture by ^{19}F NMR indicated that the Diels−Alder reaction leading to spirolactam **3a** had proceeded with high enantioselectivity (95% ee). The absolute stereochemistry was established by anamolous dispersion via X-ray crystallographic analaysis of lactam **3b** (Scheme 2 inset).

The Diels−Alder reaction leading to spirolactam **3a** with both high diastereoselectivity and enantioselectivity has a few features worthy of comment. An interesting observation is the complete unreactivity of diene (*Z*)-**5**. In contrast, we previously determined that both geometric isomers of diene **5** could be used to provide the *same diastereomer* in high yield using Et_2AlCl as promoter.[12a] These results suggest either different mechanisms for these Diels−Alder reactions (∼concerted vs completely stepwise) or the potential for olefin isomerization with the stronger Lewis acid Et_2AlCl[23] but not with the weakly Lewis acidic chiral catalyst. Calculations suggest that the energy difference between *s-cis* and *s-trans* conformers is not sufficient to rationalize the unreactivity of diene (Z)-**5** due to the low steric requirement of the alkyne.[24] In fact, the *s-cis* conformation of diene (*Z*)-**5** is lower in energy than *s-trans* (Figure 2). As expected, the

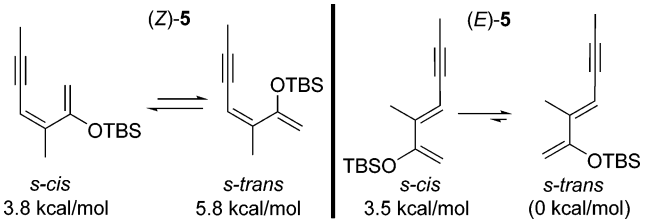


Figure 2. Calculated relative energies for dienes (*Z*)- and (*E*)-**5**.

s-trans conformer of diene (*E*)-**5** is more stable than the *s-cis* conformer but the latter conformer is accessible at room temperature. Taken together, these data suggest that the unreactivity of diene (*Z*)-**5** in the asymmetric reaction is due to steric interactions with the catalyst−ligand complex proceeding through a more concerted process (vide infra) and not inaccessibility of the reactive *s-cis* conformer.

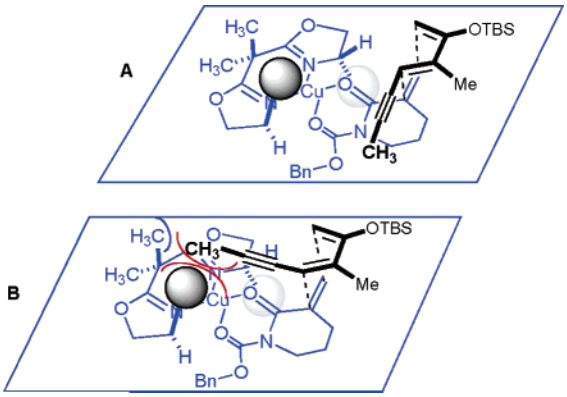


Figure 3. Favored (A) and unfavored (B) transition state arrangements that rationalize the differential reactivity of dienes (*E*) and (*Z*)-**5** in the Diels−Alder process.

Another noteworthy feature is the high diastereoselectivity resulting from an *exo*-selective reaction. This selectivity is not unusual for this type of conformationally restricted (*s-cis*), exocyclic alkene and has been attributed to favorable dipole cancellation in the *exo*-transition state.[25] In the present enantioselective process, the preference for *exo* selectivity is more pronounced due to interactions of the 2-siloxysubstituent of diene **5** with the chiral ligand in the *endo* transition state (not shown, cf. Figure 3) as previously observed by Evans in reactions of 3-methyl-1-acetoxybuadienes.[21b] The

(22) See the Supporting Information for details.

(23) We have determined that pure diene (*Z*)-**5** undergoes isomerization to a ∼1:1 mixture of geometrical isomers within ∼25 min under the Diels−Alder reaction conditions with Et_2AlCl as determined by aliquot NMR.

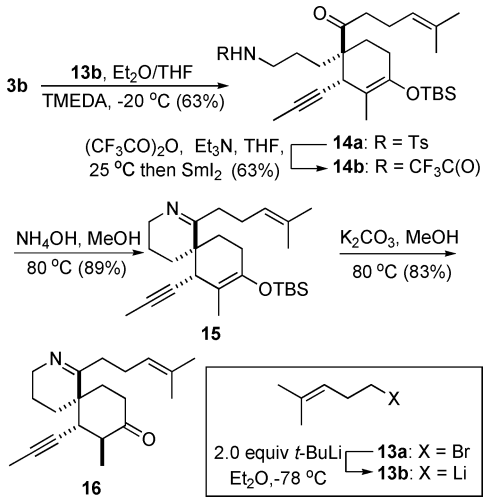
(24) DFT calculations were performed using the Gaussian 03 program package (B3LYP/6-31G* basis set + zpe). See the Supporting Information for further details.

(25) For Diels−Alder reactions with exocyclic alkenes leading to *exo* selectivity, see: (a) Roush, W. R.; Brown, B, B. *J. Org. Chem.* **1992**, *57*, 3380. (b) Fotiadu, F.; Michel, F.; Buono, G. *Tetrahedron Lett.* **1990**, *31*, 4863.

stereochemistry of adduct **3a** thus results from an *exo* trajectory of the diene (*E*)-**5** from the most accessible face opposite to the *tert*-butyl substituent (sphere) of the catalyst-dienophile complex (Figure 3A). The severe steric interaction of diene (*Z*)-**5** with the catalyst-ligand complex readily explains its low reactivity in the enantioselective Diels−Alder reaction (Figure 3B).

We previously described a one-pot Hua reaction to prepare the cyclic imine found in gymnodimine.[12b] Unfortunately, this reaction could not be translated to the real system. We ultimately found that direct addition of alkyllithiums to the *N*-tosyl lactam **3b** proceeded smoothly to provide monoalkylated products if careful attention was given to reaction temperature.[26] Thus, treatment of *N*-tosyl lactam **3b** with (4-methyl-3-pentenyl)lithium (**13b**)[27] in the presence of TMEDA at −20 °C afforded the desired amino ketone **14a** in good yield (Scheme 3). It is worth noting that the combination of an activated lactam with a reactive lithium species enabled this addition despite the proximity to a quaternary carbon center.[28]

Scheme 3. Formation of the Spirocyclic Ketoimine **16**[a]



Efforts to keep the silyl enol ether intact for the purposes of the total synthesis, led us to develop a mild, single-pot sequence to convert a *N*-tosyl group into a more labile *N*-trifluoroacetyl group. The sequence builds on related processes employed to cleave N−O bonds[29] and applied by us and others[30] to cleave *N*-benzyloxy and *N*-tosyl-*β*-lactams. Treatment of tosylamine **14a** with trifluoroacetic anhydride in the presence of base led to the formation of a putative intermediate *N*-tosyl *N*-trifluoroacetyl amide, which was subsequently reduced with samarium iodide to provide trifluoroacetamide **14b**. Cleavage of the trifluoroacetamide with warm ammonium hydroxide led to cleavage of the trifluoroacetyl group and concomitant cyclization to imine **15**. The mildness of this deprotection method makes this an attractive procedure for cleavage of primary *N*-tosyl amides. Desilylation of the silylenol ether under basic conditions provided spirocyclic ketoimine **16**. The keto group present in this spirocycle will enable linker attachment (e.g., oxime or enamine formation) and allow coupling to a carrier protein for antibody production.

In conclusion, we have completed an efficient synthesis of the spirocyclic imine moiety of gymnodimine via a highly diastereo- and enantioselective Diels−Alder reaction. Subsequent selective monoalkylation of a derived *N*-tosyl lactam with an alkyllithium furnished a ketosulfonamide precursor to the cyclic imine. A mild deprotection, cyclization sequence was developed to convert the sulfonamide to the cyclic imine. Desilylation provided a ketone readied for conjugation and evaluation as a possible immunogen for development of an immunoassay for gymnodimine and congeners. These studies and efforts toward the total synthesis will be the subject of future reports.

Acknowledgment. We thank the NIH (GM52964), the Welch Foundation (A-1280), and Pfizer for support of these investigations. We thank Dr. Nattamai Bhuvanesh (TAMU) for obtaining the X-ray structure of lactam **3b**. We thank Dr. Huda Henry-Riyad for performing the DFT calculations and Dr. Lisa Perez in the Laboratory for Molecular Simulation (TAMU) for assistance.

Supporting Information Available: Experimental procedures and characterization data for compounds **3a**, **3b**, (*E*)-**5**, **7**, **10**, **11**, **12**, **14a**, **14b**, **15**, **16**. This material is available free of charge via the Internet at http://pubs.acs.org.

OL051840R

(26) The absence of dialkylated adducts is unexpected as addition of alkyllithiums to simple α,α′-dimethyl-*N*-tosyllactams gave predominantly alcohols resulting from double addition even when 1 equiv of alkyllithium was employed (−78 → +23 °C).

(27) Hudlicky, T.; Govindan, S. V.; Frazier, J. *J. Org. Chem.* **1985**, *50*, 4166.

(28) For related examples of ring cleavage reactions of *N*-protected lactams with nucleophiles, see: (a) Flynn, D. L.; Zelle, R. E.; Grieco, P. A. *J. Org. Chem.* **1983**, *48*, 2424. (b) Giovannini, A.; Savoia, D.; Umani-Ronchi, A. *J. Org. Chem.* **1989**, *54*, 228.

(29) Keck, G. E.; McHardy, S. F.; Wager, T. T. *Tetrahedron Lett.* **1995**, *36*, 7418.

(30) (a) Rzasa, R. M.; Shea, H. A.; Romo, D. *J. Am. Chem. Soc.* **1998**, *120*, 591. (b) Taggi, A. E.; Hafez, A. M.; Wack, H.; Young, B.; Drury, W. J.; Lectka, T. *J. Am. Chem. Soc.* **2000**, *122*, 7831.