Asymmetric synthesis of batrachotoxin: Enantiomeric toxins show functional divergence against Na$_v$s

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Date: 2016/12/19

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◆ Developing tools for chemical synthesis and exploits such inventions to facilitate access to architecturally complex natural products;
◆ Contemporaneous efforts to explore the mechanism of C–H amination and to evolve new catalytic systems for C–C, C–N, and C–O bond formation.

Education:

■ B.S. University of California, Berkeley, 1992;

Research experience:

■ Postdoctoral Fellow
  Massachusetts Institute of Technology (Prof. Stephen Lippard), 1997-1999;
■ Independent Research
  Department of Chemical & Systems Biology, Stanford University, (1999-now).
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☑ Summary
An extremely potent cardiotoxic and neurotoxic steroidal alkaloid found in certain species of frogs (poison dart frog), melyrid beetles, and birds;

Neurotoxin, irreversibly binds to the Na\(^+\) channels to keep it open, LD\(_{50}\) in mice: 2 ug/kg, in comparison, LD\(_{50}\) (NaCN) in mice: 6.3 mg/kg;

Structural features: a pentacyclic core skeleton, an intramolecular hemiketal, a seven membered oxazapane ring.
Formal synthesis of (+/−)-Batrachotoxin

Retrosynthetic Analysis

Synthesis (+/-)-cis-Decalone 13

\[ \text{Synthesis (+/-)-cis-Decalone 13} \]

1. **5** + **6** \(\rightarrow\) EtONa, EtOH \(\rightarrow\) **7**
   - Yield: 95%
   - Reagents: **NaBH\(_4\)**, EtOH, 0 °C, \(\rightarrow\) **8**
   - Yield: 87% (2 steps)

2. **9** \(\rightarrow\) 2-Napthalene sulfonylic acid
   - Yield: 75%
   - Reagents: \(\text{Na}_2\text{CO}_3\cdot 1.5\text{H}_2\text{O}_2\), TFAA
   - Yield: 87%
   - Reagents: 1) 5% HCl, 2) DBU
   - Yield: 95% (2 steps)

3. **11** \(\rightarrow\) 1) Pd/C, H\(_2\), 2) Me\(_4\)NB(OAc)\(_3\)H
   - Yield: 76% (2 steps)
   - Reagents: TBSCl, imidazole
   - Yield: 97%
   - Reagents: TBSO

**13**
Synthesis of furan substrate 18

13\[\text{Me, OAc, TBS, OTBS} \xrightarrow{\text{1) DIBAL-H, -78 °C}} \xrightarrow{\text{2) DMSO, (COCl)}_2 \text{Et}_3\text{N, CH}_2\text{Cl}_2} \xrightarrow{76\% (2 steps)} 14\[\text{Me, K, TBS, OTBS} \xrightarrow{\text{1) HCO}_2\text{Et, NaH}} \xrightarrow{\text{2) }^7\text{BuSH, p-TsOH, benzene, reflux}} \text{88\% (2 steps)}

15\[\text{Me, S^Bu, TBS, OTBS} \xrightarrow{\text{1) Me}_3\text{Si, NaHMDS}} \xrightarrow{\text{2) HgCl}_2, \text{Et}_2\text{O}} \text{54\% (2 steps)}

16\[\text{Me, OTBS} \xrightarrow{\text{DMF, (COCl)}_2} \text{84\%}

17\[\text{Me, S^Bu, TBS, OTBS} \xrightarrow{\text{1) Ph}_3\text{P=CHOME}} \xrightarrow{\text{2) 1,3-propanedithiol, CSA}} \text{72\% (2 steps)}

18\[\text{Me, S^Bu, TBS, OTBS}

Synthesis of building block 3

1) iBuLi, HMPA, 19
2) TBAF

52% (2 steps)

1) MnO2
2) NaBH3CN, 20
3) Ac2O, pyridine

75% (3 steps)

1) PIFA, MeOH
2) PPTS, acetone
3) DBU

68% (3 steps)

1) p-Nitroperoxy benzoic acid
2) MOMCl, DIEA

84% (2 steps)

1) KHMDS, 24
2) TFAA, DMSO, TEA

82% (2 steps)

1) (Me2N)3S(Me3SiF2)
2) PhNTf2, TEA

95% (2 steps)
Synthesis of building block 30

1) PTIO₂, H₂, 2) NaBH₄

1) TBAF, 2) Dess-Martin

90% (4 steps)

1) DBU, 2) CSA, MeOH

85%

1) NaBH₄, CeCl₃, 2) 2,2'-dipyridyl disulfide (n-Bu)₃P

1) KHMDS, PhNTf₂

73% (4 steps)

86% (2 steps)
Synthesis of (+/-)-Batrachotoxin 1

31 \( R = 1\)-morpholine 86% (2 steps)

32 66% (2 steps)

BTX-A 2

(+/−)-Batrachotoxin 1
Asymmetric synthesis of (-)-Batrachotoxin

Retrosynthetic Analysis of (-)-Batrachotoxin 1

Synthesis of building block 5

\[ \text{CHO} + \text{CO}_2\text{Et} \xrightarrow{\text{K}_2\text{CO}_3, \text{H}_2\text{O}} \text{EtO}_2\text{C} \xrightarrow{\text{Lipase PS, TBME, } 30 \, ^\circ\text{C}} \text{EtO}_2\text{C} \]

\[ (+/\text{-})-9 \rightarrow 48\%/48\% \rightarrow 10 \, (R = \text{Ac}) \]

\[ (R)-9 \, (R = \text{H}) \xrightarrow{\text{K}_2\text{CO}_3} (S)-9 \]

\[ \text{EtO}_2\text{C} \xrightarrow{\text{TBSCl, Et}_3\text{N, DMAP, DCM}} \text{EtO}_2\text{C} \xrightarrow{\text{DIBAL-H, Tol., } -78 \, ^\circ\text{C}} \text{EtO}_2\text{C} \]

\[ (S)-9 \rightarrow 89\% \rightarrow 11 \rightarrow 75\% \rightarrow 12 \rightarrow 96\% \]

\[ \text{I}_2, \text{PPh}_3, \text{Imidazole} \xrightarrow{} \text{TBSO} \xrightarrow{\text{Zn, AcOH}} \text{TBSO} \xrightarrow{\text{IBX}} \text{TBSO} \]

\[ 13 \rightarrow 90\% \rightarrow 14 \rightarrow 99\% \rightarrow 15 \rightarrow 92\% \rightarrow 5 \]
Synthesis of building block 6

1. L-Proline, DMF, 16 °C
2. H$_2$SO$_4$, 95 °C

90% (2 steps)

17

Pd/C, H$_2$
then HCl
ethylene glycol

89%

18

TESOTf, Et$_3$N

88%

19

CHBr$_3$
KO'Bu, -25 °C

53%

20

TMSC≡CLi
THF, 0 °C

80%

21

CSA, MeOH

91%

6

MeO
Construction of C ring via radical cascade

1. Reaction of 6 with 5 under iBuLi, -90 °C, then 5 to give 22 in 65% yield.
2. Treatment of 22 with K₂CO₃, MeOH followed by [Si]Cl, imidazole to give 24 in 87% yield (2 steps).

Steps:
- Reaction of 6 with 5 under iBuLi, -90 °C, then 5 to give 22 in 65% yield.
- Treatment of 22 with K₂CO₃, MeOH followed by [Si]Cl, imidazole to give 24 in 87% yield (2 steps).
Construction of building block 29

1. \(\text{Bu}_3\text{Sn}\) \(\text{Me}\) \(\text{OH}\) \(\text{Bu}_3\text{Sn}\) \(\text{Me}\) \(\text{OH}\)

   2-iodoxybenzoic acid
   \(^t\text{BuOH}, 65 ^\circ\text{C}\)
   then \(\text{OsO}_4, \text{NaI}_2\text{O}_4\)

   \(\text{MeO}\) \(\text{Me}\) \(\text{MeO}\) \(\text{Me}\)

   94%

2. \(\text{OH}\) \(\text{CHO}\) \(\text{MeNH}_2, \text{CH}_2\text{Cl}_2, \text{NaB(TFA)}_3\text{H}, -78 ^\circ\text{C}\)
   then \(\text{ClCH}_2\text{COCl}\)

   \(\text{MeO}\) \(\text{Me}\) \(\text{MeO}\) \(\text{Me}\)

   54%

3. \(\text{Cl}\) \(\text{MeN}\) \(\text{Me}\) \(\text{Bu}_3\text{Sn}\) \(\text{Me}\) \(\text{OH}\)

   \(\text{NaOEt, EtOH}\)
   \(\text{THF/C}_6\text{H}_6\ 1:1\)

   \(\text{Bu}_3\text{Sn}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{OH}\)

   92%

4. \(\text{Me}\) \(\text{N}\) \(\text{O}\) \(\text{Bu}_3\text{Sn}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{OTf}\)

   \(\text{KN(TMS)}_2, \text{PhNTf}_2\)
   \(\text{THF, -78 } ^\circ\text{C} \text{ to } 0 ^\circ\text{C}\)

   \(\text{Bu}_3\text{Sn}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{OTf}\)

   94%

5. \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{OTf}\)

   \(\text{CuCl}_2, \text{O}_2\)
   1,4-dioxane

   \(\text{OHC}\) \(\text{O}\) \(\text{O}\)

   85%

6. \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{O}\)

   \(\text{NaClO}_2, \text{NaH}_2\text{PO}_4, \text{DMSO/H}_2\text{O}\)

   \(\text{MeO}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\) \(\text{Me}\)
Construction of building block 35

1) SOCl₂, pyridine
2) NaN₃, acetone/H₂O

aq. AcOH, 90 °C
Curtius degradation

57% (4 steps)

30

31

32

33

p-TsOH, 4 Å MS
PMBCH₂Cl, C₆H₆

PMBH₂CO

89%

34

AlH₃, THF

33%

tributyl(1-ethoxyvinyl)tin

35

tributyl(1-ethoxyvinyl)tin

77%
Completion of synthesis of Batrachotoxin

\[
\text{PMBH}_2\text{CO}\quad 35 \quad \xrightarrow{p-\text{TsOH}} \quad \text{acetone/H}_2\text{O 3:2} \quad \xrightarrow{83\%} \quad \text{BTX-A 2}
\]

\[
\text{BTX-A 2} + \text{EtO} \xrightarrow{\text{Et}_3\text{N, C}_6\text{H}_6, 45 \degree\text{C}} \xrightarrow{70\%} \text{(-)-Batrachotoxin 1}
\]
Summary

**Batrachotoxin**

**Kishi's work**

[I] Racemic formal synthesis, 40 steps

[II] [4+2] Cycloaddition for constructing C&D rings

[III] Intramolecular oxa-Michael addition for E ring

**Du Bois's work**

[I] Asymmetric total synthesis, 26 steps

[II] Radical cascade for constructing C&D rings

[III] Intramolecular S_N2 reaction for E ring
The phenotypic effects of acute poisons found among the rich pharmacopeia of terrestrial and marine life have been documented from antiquity. Isolation and characterization of toxic compounds have made available important chemical reagents for studying complex biochemical circuits. Studies of this type have revealed a large number of peptide and small molecule agents that target voltage-gated sodium ion channels (NaVs), an obligatory class of membrane proteins for bioelectrical signaling. Among the collection of known NaV modulators are three structurally related agents, (−)-batrachotoxin, veratridine, and aconitine, sterically large, lipophilic amine derivatives believed to share a common binding locus in the inner pore region of NaV.
The influence of these toxins on ion gating, however, differs distinctly. On one extreme, (−)-BTX, the primary toxic constituent of Colombian poison dart frogs, is a full Naᵥ agonist, causing the channel to open more readily at hyperpolarized membrane potentials and blocking fast inactivation. Conversely, the activities of veratridine and aconitine are best described as partial agonism and inhibition of channel function, respectively. Despite recent insights from structural biology into the three-dimensional architecture of prokaryotic Naᵥ, a molecular understanding of the influence of the site toxins on ion conduction and ion gating kinetics is lacking.
Toxin structure-activity studies, in combination with protein mutagenesis experiments, can address questions related to the dynamical nature of channel function and may guide the rational design of small-molecule modulators of Na\textsubscript{V} activity. The potency of (−)-BTX, its storied history as the archetypal small-molecule site probe, and its unparalleled effects on channel gating render it an optimal “lead” compound for such investigations.