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Total synthesis of (+)-kalkitoxin†

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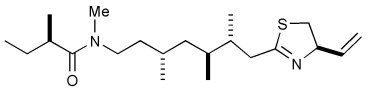
The neurotoxic lipopeptide (+)-kalkitoxin was synthesized by a route which employed asymmetric organocopper conjugate addition followed by *in situ* enolate alkylation to install the *anti*,*anti*-1,2,4-trimethyl relationship of the toxin; the synthesis of kalkitoxin required sixteen steps and proceeded in 3% overall yield.

The lipopeptide kalkitoxin (**1**) was isolated from an extract of the cyanobacterium *Lyngbya majuscula* collected in the Caribbean[1] and has been shown to possess potent neurotoxicity in a primary cell culture of rat neurons (LC_{50} 3.86 nM).[2] Kalkitoxin is also highly active in an inflammatory disease model which measures IL-11b-induced $sPLA_2$ secretion from HepG2 cells (IC_{50} 27 nM),[3] and there is evidence to suggest that **1** is a potent blocker of the voltage sensitive sodium ion channel in mouse neuro-2a cells (EC_{50} 1 nM; *cf.* EC_{50} of saxitoxin is 8 nM).[4]



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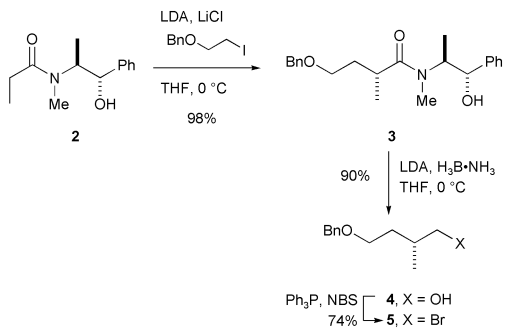
These properties make kalkitoxin and analogues attractive targets for synthesis, and we report herein a concise route to the natural (3*R*,7*R*,8*S*,10*S*,2′*R*) configuration of **1** employing a tandem organocopper conjugate addition-alkylation sequence for installing the 1,2,4-*anti*,*anti*-trimethyl relationship of the toxin. This strategy diverges from a previous route to **1** used to establish the structure and absolute configuration of kalkitoxin in which the C7, C8, and C10 methyl substituents were introduced sequentially.[1]

Our synthesis of **1** commenced from the lithium enolate of (1*S*,2*S*)-*N*-propionylpseudoephedrine (**2**)[5] which was alkylated with 2-iodoethyl benzyl ether[6] (Scheme 1). The resulting amide **3** was cleaved reductively with lithium amidotrihydroborate[7] to yield alcohol **4**. The latter was converted to bromide **5** which, after transformation to the corresponding organocopper species,



Scheme 1

† Electronic supplementary information (ESI) available: experimental procedures and spectroscopic data. See http://www.rsc.org/suppdata/cc/b3/b306124h/

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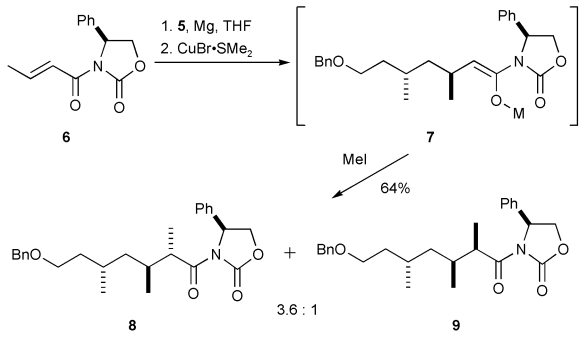


Scheme 2

was reacted with (*S*)-*N*-(*trans*-crotonyl)-4-phenyloxazolidin-2-one (**6**)[8] (Scheme 2).

The *anti*-1,3-dimethyl array of enolate **7** generated from **5** and **6** represents a matched case and has precedent in studies by Williams[9] who has proposed a transition state model for this process.[10] The use of Hruby's 4-phenyloxazolidinone auxiliary is of crucial importance in this asymmetric conjugate addition since the Evans auxiliary, in which the phenyl substituent is replaced by benzyl, gave significantly lower asymmetric induction. Quenching of **7** and subsequent methylation of the enolate in a separate step was found to yield a mixture of **8** and **9** favoring the former (2.8 : 1), but slightly improved stereoselectivity occurred (3.6 : 1) when enolate **7** from organocopper conjugate addition to **6** was trapped directly with methyl iodide. After chromatographic separation from its isomer **9**, oxazolidinone **8** was crystallized and its structure was confirmed by X-ray crystallographic analysis (Fig. 1).‡

Reductive removal of the benzyl ether from **8** and Swern oxidation of the resultant primary alcohol gave aldehyde **10**, which was subjected to reductive amination with methylamine and sodium cyanoborohydride (Scheme 3). The resulting secondary amine **11** was coupled to (*R*)-2-methylbutyric acid[11] in a process that required HOAt[12] as an activating agent to yield amide **12** (mixture of rotamers). Reductive cleavage of the chiral adjuvant with lithium borohydride then furnished primary alcohol **13**. This substance required a one-carbon homologation before proceeding toward construction of the thiazoline moiety of **1**, and for this purpose, **13** was oxidized to the corresponding aldehyde. The latter was subjected to Wittig olefination with the ylide from methoxymethyltriphenylphosphonium chloride[13] to furnish a mixture of (*E*)- and (*Z*)-enol ethers **14**. Hydrolysis of

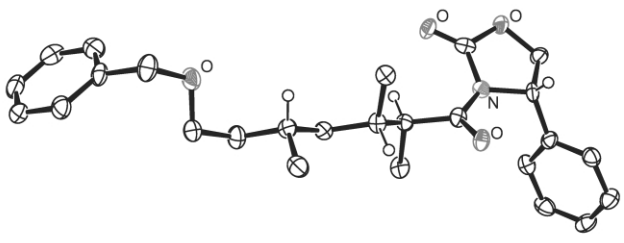
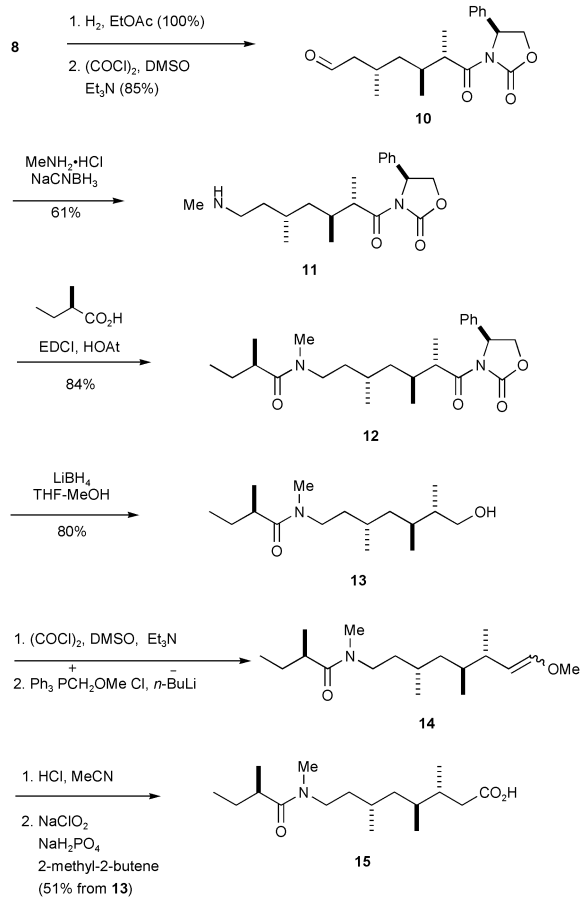


Fig. 1 X-Ray crystal structure of **8** (ORTEP at 50% probability level).

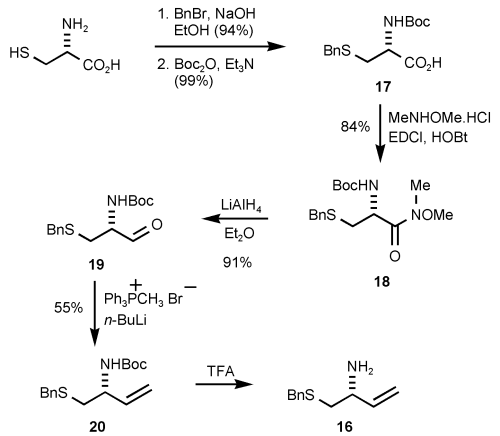


Scheme 3

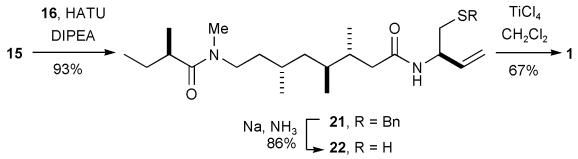
14 and oxidation of the resulting aldehyde produced carboxylic acid **15**.

For synthesis of the thiazoline portion of kalkitoxin we employed a route based on titanium tetrachloride-mediated cyclization of an amido thiol.[14,15] Thus, (*R*)-3-amino-4-thiobenzylbut-1-ene (**16**) required for coupling with **15** was prepared from (*R*)-cysteine which was first protected as its *N*-Boc-*S*-benzyl derivative **17** (Scheme 4).[16]

This carboxylic acid was converted to its Weinreb amide **18**,[17] reduction of which gave aldehyde **19**. Wittig methylenation of **19** produced **20**, and subsequent removal of the Boc protection afforded **16**. Coupling of this amine with carboxylic acid **15**, using HATU for activation,[18] led to bis-amide **21** in



Scheme 4



Scheme 5

excellent yield, and subsequent reductive cleavage of the benzyl thioether with sodium–ammonia furnished thiol **22** (Scheme 5). Finally, exposure of **22** to titanium tetrachloride yielded kalkitoxin (**1**, $[\alpha]_D^{20}$ + 11.5 (*c* 0.34, $CHCl_3$)), whose ^1H and ^{13}C NMR spectra precisely matched those of the natural material.

The sixteen-step sequence to kalkitoxin described above proceeds in *ca.* 3% overall yield (Scheme 5) and has provided sufficient material for comprehensive studies of neurotoxic and cytotoxic properties. The results of these studies will be published in due course.

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Notes and references

‡ *Crystal data* for **8**: M = 425.53; monoclinic, space group $C2$ (no. 5); a = 29.048(7), b = 5.7108(6), c = 14.709(3) Å, β = 102.212(8)°, V = 2384.8(8) Å^3, T = 93(2) K; Z = 4; μ(Cu-Kα) = 0.629 mm^{-1}; reflections: total = 3851, unique = 2613 (R_{int} = 0.0317); residuals (all data, Shelxl): $R1$ = 0.0862, $wR2$ = 0.1453; Absolute structure parameter (Flack) = 0.1(6) (the absolute structure cannot be uniquely determined based on diffraction data alone). CCDC 211921. See http://www.rsc.org/suppdata/cc/b3/b306124h/ for crystallographic data in CIF or other electronic format.

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