Tetrahedron 65 (2009) 4007–4012

Contents lists available at ScienceDirect

Tetrahedron

journal homepage: www.elsevier.com/locate/tet

Total synthesis and confirmation of the absolute stereochemistry of semiviriditoxin, a naphthopyranone metabolite from the fungus *Paecilomyces variotii*

Nichole P.H. Tan [a,b], Christopher D. Donner [a,b,*]

[a] School of Chemistry, The University of Melbourne, Victoria 3010, Australia
[b] Bio21 Molecular Science and Biotechnology Institute, The University of Melbourne, Victoria 3010, Australia

ARTICLE INFO

Article history:
Received 30 November 2008
Received in revised form 16 February 2009
Accepted 6 March 2009
Available online 11 March 2009

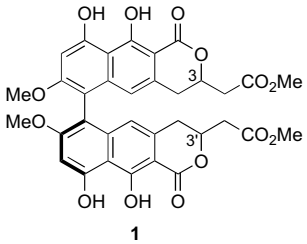
ABSTRACT

The first total synthesis of (*S*)-semiviriditoxin **2** is described. The approach utilizes a tandem Michael–Dieckmann reaction between *ortho*-toluate **5** and dihydropyran-2-one **6** to construct the naphthopyranone core, the dihydropyran-2-one **6** being prepared from (*R*)-1,2-epoxy-4-butanol. Spectroscopic comparison of synthetic (*S*)-semiviriditoxin **2** with (*R*)-semivioxanthin **3**, prepared in four steps from (*R*)-propylene oxide, confirmed the (*S*)-stereochemistry of natural semiviriditoxin from *Paecilomyces variotii*.

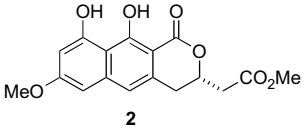
1. Introduction

The increasing occurrence of bacterial resistance to antibiotics, and its consequences for human health, necessitates the development of new antibiotics that possess novel modes of action. The bacterial protein FtsZ, a tubulin-like GTPase, has an essential role in bacterial cell division wherein it undergoes self-polymerization, thus initiating the complex process of septation. The high degree of similarity of FtsZ amongst bacterial species makes it an excellent target for antibiotic drugs.[1] Viriditoxin **1** has been shown to block FtsZ polymerization with an IC_{50} of 8.2 μg/mL, and possesses broad-spectrum antibiotic activity against Gram-positive pathogens, including methicillin-resistant *Staphylococcus aureus* and vancomycin-resistant *Enterococci*.[2] First isolated in 1971 from *Aspergillus viridinutans* the structure of viriditoxin was originally incorrectly assigned,[3] and has since been revised to the structure **1**.[4] Whilst the chirality of the biaryl axis in **1** was established from the circular dichroism (CD) spectrum, the stereochemistry at C3 and C3′ in viriditoxin is not known. Semiviriditoxin **2**, the monomeric subunit of **1**, has been obtained from the fungus *Paecilomyces variotii*.[5] The similarity of the CD spectrum of **2** with that of (*R*)-semivioxanthin **3** led to the proposal of (*S*)-stereochemistry for semiviriditoxin **2**. The closely related dimeric naphthopyranone asteromine **4** has been isolated from a strain of *Mycosphaerella asteroma*,[6] however, **4** exhibits only weak antibacterial and antifungal activities.

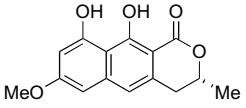
In order to ultimately solve the outstanding stereochemical details of viriditoxin **1**, we sought to undertake a stereospecific synthesis of the potential precursor semiviriditoxin **2**, a natural product that itself has yet to be the subject of total synthesis. A concise method for the preparation of naphthopyranones utilizes the tandem Michael–Dieckmann reaction of *ortho*-toluates with 4-alkoxypyran-2-ones, a method first reported by Staunton and co-workers.[7] Such an approach has been used in both racemic[8] and enantioselective[9] syntheses of semivioxanthin **3**. We have used this methodology towards the synthesis of the talaroderxines[10] and as the key step in the preparation of pyranonaphthoquinones.[11] Herein we report the application of this method to both the first total synthesis of (*S*)-semiviriditoxin **2** and a concise synthesis of (*R*)-semivioxanthin **3**, allowing the absolute stereochemistry of natural semiviriditoxin **2** to be firmly established.



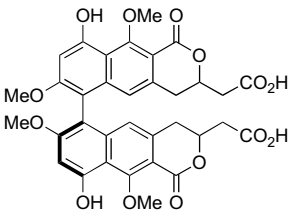
1



2



3



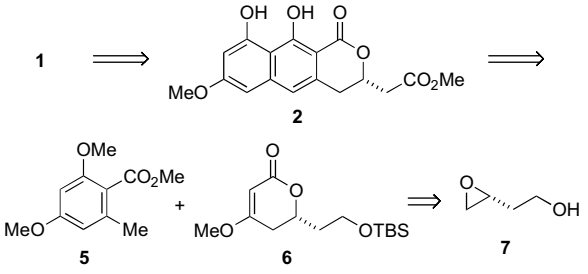
4

* Corresponding author. Tel.: +61 3 8344 2411; fax: +61 3 9347 8189.
E-mail address: cdonner@unimelb.edu.au (C.D. Donner).

0040-4020/$ – see front matter
doi:10.1016/j.tet.2009.03.016

2. Results and discussion

The symmetrical nature of viriditoxin **1** would logically suggest the potential for synthesis via oxidative phenolic coupling of the corresponding monomer semiviriditoxin **2** (Scheme 1). The full carbon framework of **2** should be amenable to rapid construction utilizing a tandem Michael–Dieckmann reaction between the *ortho*-toluate **5** and lactone **6**. In turn, lactone **6** should be available from the terminal epoxide **7**, which itself is available from (*S*)-aspartic acid.[12]



Scheme 1.

Firstly, (*S*)-aspartic acid **8** was converted to the epoxyalcohol **7** in 85% yield over three steps following the method of Volkmann and co-workers (Scheme 2).[12] Silylation of the alcohol **7** proceeded efficiently to give the protected (*R*)-epoxide **9**, the spectroscopic data of which (see Experimental) were in good agreement with that reported for the same material prepared from (*S*)-malic acid.[13] The epoxide **9** was then treated with the anion of methyl propiolate under the Yamaguchi–Hirao conditions[14] to deliver the acetylene **10** in 72% yield. Exposure of the acetylene **10** to sodium methoxide (0.3 equiv) in methanol[15] leads to conjugate addition and subsequent lactonization to give the pyranone **6** in 80% yield. The initial adduct formed between **10** and methanol would presumably be formed as a mixture of isomers,[16] however, under the reaction conditions equilibration between the *E*/*Z* isomers can occur. Ultimately, all the *E*-isomer is trapped as the lactone **6** and no uncyclized material was observed. With the key stereochemically defined lactone fragment in hand, we could then proceed with the tandem Michael–Dieckmann reaction.

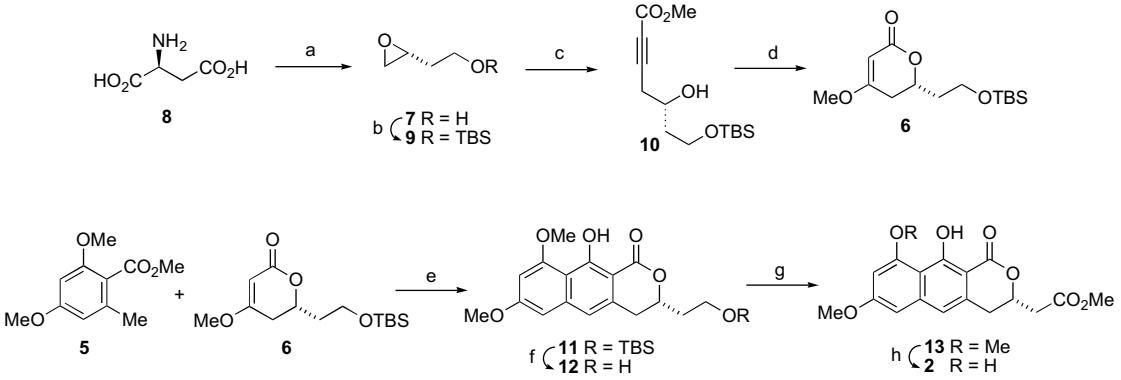
We have recently investigated the scope of the tandem Michael–Dieckmann reaction between oxygenated *ortho*-toluates and 4-methoxypyran-2-ones.[17] Fortuitously in the present context, the toluate **5** is amenable to reaction with the lactone **6**. Thus, deprotonation of the toluate **5** with LDA at −65 °C followed by addition of lactone **6** delivers the naphthopyranone **11** in 36% yield. The ^{1}H NMR spectrum of **11** shows signals consistent with the assigned structure, including three aromatic signals at δ 6.47 (d, *J* 2.2 Hz), 6.58 (d, *J* 2.2 Hz), and 6.85 (s) and a chelated phenolic proton signal at δ 13.14. The naphthopyranone **11** contains the complete carbon framework found in semiviriditoxin **2**, only requiring seemingly trivial functional group manipulations for its ultimate conversion to **2**.

The silyl group in **11** was removed by exposure to aq HCl/THF to deliver the primary alcohol **12** in 70% yield after recrystallization. The analytical and spectroscopic data (Experimental) were in complete accord with structure **12**. The primary alcohol **12** was then converted to the corresponding methyl ester **13** by sequential oxidation (TEMPO/PhI(OAc)$_2$ then $NaClO_2$) followed by esterification (MeOH, concd H_2SO_4) to give semiviriditoxin-9-*O*-methyl ether **13** in 84% yield for the three steps. As anticipated, the spectroscopic data for **13** were very similar to that reported[5] for semiviriditoxin **2**. Notably, the ^{1}H NMR spectrum of **13** differs only by the presence of an extra methoxy signal (δ 3.98), replacing the weakly chelated proton signal (δ 9.44) seen in the spectrum of **2**.

Final conversion of **13** to semiviriditoxin **2** requires selective demethylation of the C9 methyl ether. The use of BCl_3 gave the best results for this conversion, the reaction being stopped before complete conversion to ensure that disruption of the C7 methyl ether was avoided. After separation from unreacted **13**, (*S*)-semiviriditoxin **2** was obtained as a colourless solid, mp 169–171 °C (lit.[5] mp 168–170 °C), with both the ^{1}H and ^{13}C NMR spectra for the synthetic material being virtually identical to the reported data for the natural product.[5] Significantly, the specific rotation for (*S*)-semiviriditoxin **2** prepared in this way, $[\alpha]_D^{23}$ −8.60 (*c* 0.06, $CHCl_3$), closely matches the reported value for semiviriditoxin, $[\alpha]_D^{20}$ −9.0 (*c* 0.21, $CHCl_3$),[5] and on this basis the previously proposed (*S*)-stereochemistry for semiviriditoxin **2** from *P. variotii* can be verified.

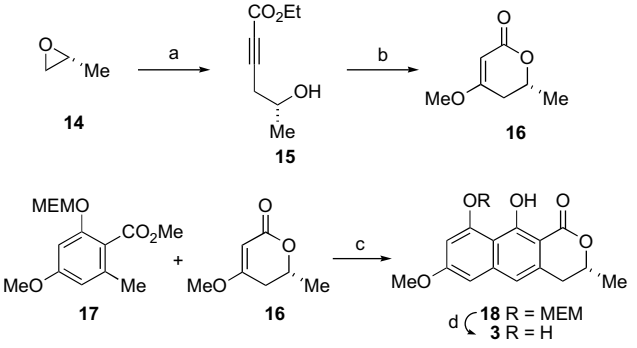
Although the similarity in specific rotation values between synthetic **2** and that reported for the naturally occurring material seemed to confirm the (*S*)-stereochemistry of natural semiviriditoxin **2**, we thought it prudent to corroborate this conclusion. Previously, the (*S*)-stereochemistry of semiviriditoxin from *P. variotii* was proposed based on the similarity of the CD spectrum of **2**[5] with the reported CD data for (*R*)-semivioxanthin **3** from *Penicillium citreo-viride*.[18,19] Given that the CD data for semiviriditoxin **2** and semivioxanthin **3** were obtained in different solvents, we felt that direct comparison of these materials under similar conditions would be warranted in order to confirm the legitimacy, in this case, of assigning the stereochemistry in this manner.

To obtain (*R*)-semivioxanthin **3** we undertook the concise synthesis outlined in Scheme 3. The approach closely parallels that of Drochner and Müller for their synthesis of (*R*)-semivioxanthin.[9] In



Scheme 2. Reagents and conditions: (a) (i) $NaNO_2$, H_2SO_4, KBr, H_2O, 0 °C, 4 h (91%); (ii) $BH_3{\cdot}Me_2S$, THF, −30 °C to rt, 18 h (97%); (iii) K_2CO_3, CH_2Cl_2, 72 h (96%); (b) TBSCl, imidazole, CH_2Cl_2, 4 h (98%); (c) methyl propiolate, ^{n}BuLi, $BF_3{\cdot}Et_2O$, THF, −78 °C, 30 min (72%); (d) NaOMe, MeOH, 16 h (80%); (e) LDA, THF, −60 °C, 30 min (36%); (f) THF, 10% HCl, 16 h (70%); (g) (i) PhI(OAc)$_2$, TEMPO, CH_2Cl_2, 20 h; (ii) $NaClO_2$, ^{t}BuOH, H_2O, NaH_2PO_4, 3 h; (iii) MeOH, concd H_2SO_4, 16 h (84%, three steps); (h) BCl_3, CH_2Cl_2, 6 h (75%).

this previously reported synthesis of **3**, the lactone **16** was obtained in three steps from *tert*-butyl 3,5-dioxohexanoate using a regio- and stereoselective (99.4% ee) enzymatic reduction, lactonization, and O-methylation sequence, whilst a benzyloxymethyl protected *ortho*-toluate was employed in the tandem reaction. In the present work we prepared the (*R*)-lactone **16** in just two steps from (*R*)-propylene oxide **14**. Thus, addition of the (*R*)-epoxide **14** to the anion of ethyl propiolate gave the acetylene **15** (Scheme 3), which upon exposure to catalytic sodium methoxide gave the lactone **16** in 66% yield over the two steps. In the present case we chose to use the methoxyethoxymethyl (MEM) protected toluate **17** for the tandem Michael–Dieckmann reaction with (*R*)-lactone **16**, delivering the naphthopyranone **18** in 39% yield. Final deprotection of **18** provided semivioxanthin **3** in 17% yield over the four steps. This compares favourably with the method of Drochner and Müller that produced **3** in 5% yield over five steps. Importantly, the spectroscopic data for synthetic (*R*)-semivioxanthin **3** obtained in this way were in complete agreement with the reported data for naturally occurring semivioxanthin **3**.[18]



Scheme 3. Reagents and conditions: (a) ethyl propiolate, ^{n}BuLi, $BF_3\cdot Et_2O$, THF, −90 °C, 2 h (82%); (b) NaOMe, MeOH, 16 h (80%); (c) LDA, THF, −60 °C, 30 min (39%); (d) THF, concd HCl, 8 h (66%).

With both (*S*)-semiviriditoxin **2** and (*R*)-semivioxanthin **3** now available to us we could undertake their direct spectroscopic comparison. The CD spectrum of synthetic (*S*)-semiviriditoxin **2** (obtained in trifluoroethanol) exhibited a maximum at 370 nm and minimum at 262 nm, in close agreement with the reported data for natural semiviriditoxin (in dioxane),[5] whilst synthetic (*R*)-semivioxanthin **3** (in trifluoroethanol) exhibited a maximum at 363 nm and minimum at 266 nm, also in accord with reported data for natural semivioxanthin (in cyclohexane/5% dioxane).[18] Furthermore, the near identity of the CD spectra for synthetic **2** and **3**, obtained here under similar conditions, supports the previous application of this technique for the assignment of the stereochemistry of semiviriditoxin.[5]

In summary, this work constitutes the first total synthesis of the fungal metabolite (*S*)-semiviriditoxin **2**. Spectroscopic comparison of synthetic **2** with the reported data for semiviriditoxin **2** and with synthetic (*R*)-semivioxanthin **3**, prepared in four steps from (*R*)-propylene oxide, has confirmed the (*S*)-stereochemistry of semiviriditoxin from *P. variotii*. At this stage, attempted dimerization of **2** to give viriditoxin **1** has not been successful and investigations into methods to effect this transformation are ongoing.

3. Experimental section

3.1. General

All moisture sensitive reactions were performed under a dry nitrogen atmosphere in oven-dried or flame-dried glassware. Thin layer chromatography (TLC) was performed on pre-coated silica plates (Merck $60GF_{254}$) and compounds were visualized at 254 nm and 365 nm or stained with 20% w/w phosphomolybdic acid in ethanol. Flash column chromatography was performed on silica gel (Kieselgel 60, 230–400 mesh) using the indicated solvent system. Melting points were measured using a Bausch and Lomb hot-stage melting point apparatus and are uncorrected. ^{1}H and ^{13}C NMR spectra were recorded using a Varian-500 spectrometer operating at 500 MHz and 125 MHz, respectively. The residual peak of $CHCl_3$ was used as internal reference (7.26 ppm) for ^{1}H NMR spectra whilst the central peak of $CDCl_3$ was used as reference (77.0 ppm) for ^{13}C NMR spectra. Chemical shifts are reported in parts per million (ppm). ^{1}H NMR data are reported in the following order: number of protons, multiplicity, coupling constant (*J*, Hz), assignment. Infrared (IR) spectra were recorded on a Perkin–Elmer Spectrum One FT-IR spectrometer. Low resolution electrospray ionization (ESI) mass spectra were recorded on a Shimadzu GC/MS-QP505A. High resolution ESI mass spectra were recorded on a Thermo-Finnigan LTQ-FT ICR hybrid mass spectrometer. Specific rotations were measured at 589 nm using a JASCO DIP-1000 polarimeter using a 1 dm cell with concentrations quoted in g/100 mL in the solvent cited in each case and are given in units 10^{-1} deg cm^2 g^{-1}. Circular dichroism spectra were obtained using a JASCO J-815 CD spectrometer for solutions in the solvent specified. Anhydrous tetrahydrofuran (THF) and dichloromethane were pre-dried over activated alumina under argon. Ether refers to diethyl ether, while petrol refers to the hydrocarbon fraction boiling in the range 40–60 °C. Micro elemental analyses were conducted by Chemical and MicroAnalytical Services Pty. Ltd., Geelong, Victoria, Australia.

3.2. (*R*)-2-(2-(*tert*-Butyldimethylsilyloxy)ethyl)oxirane 9

To the (*R*)-epoxide **7** (2.4 g, 27.2 mmol) in dichloromethane (50 mL) were added imidazole (1.95 g, 28.6 mmol) and *tert*-butyldimethylsilyl chloride (4.31 g, 28.6 mmol) and the mixture was stirred at ambient temperature for 4 h. Ether (75 mL) was added and the mixture was washed successively with water, dilute hydrochloric acid, 10% sodium hydrogen carbonate and brine, then dried ($MgSO_4$) and concentrated in vacuo to give the title compound **9** (5.4 g, 98%) as a colourless oil that was used without further purification. $[\alpha]_D^{24}$ +11.0 (*c* 2.00, $CHCl_3$) {lit.[13] $[\alpha]_D^{26}$ +12.5 (*c* 2.11, $CHCl_3$)}; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 0.06 (6H, s, $Si(CH_3)_2$), 0.90 (9H, s, $SiC(CH_3)_3$), 1.67–1.81 (2H, m, CH_2), 2.52 (1H, d, *J* 5.1 and 2.8 Hz, 1-H_aH_b), 2.78 (1H, m, 1-H_aH_b), 3.05 (1H, m, 2-H), 3.78 (2H, t, *J* 5.8 Hz, CH_2OSi); ^{13}C NMR ($CDCl_3$, 125 MHz) δ −5.4, 18.3, 25.9, 35.9, 47.2, 50.0, 60.0.

3.3. Methyl (*S*)-7-(*tert*-butyldimethylsilyloxy)-5-hydroxyhept-2-ynoate 10

To a solution of methyl propiolate (0.41 g, 4.82 mmol) in THF (8 mL) at −78 °C was added *n*-butyllithium (2.5 M in hexane, 1.93 mL, 4.82 mmol) dropwise and the mixture was stirred for 10 min at −78 °C. Boron trifluoride/diethyl etherate (0.65 mL, 5.14 mmol) was added and stirring was continued for 10 min. A solution of (*R*)-epoxide **9** (0.65 g, 3.21 mmol) in THF (1 mL) was added and stirring was continued for 30 min at −78 °C. Aqueous ammonium chloride was added and the mixture was extracted with ethyl acetate (4×5 mL), the combined organic extracts washed with brine, dried ($MgSO_4$) and concentrated in vacuo. Flash column chromatography (ethyl acetate/petrol 1:3) afforded the title compound **10** (0.66 g, 72%) as a colourless oil. $[\alpha]_D^{26}$ +3.10 (*c* 2.00, $CHCl_3$). Found: $[M+Na]^+$ 309.1492. $C_{14}H_{26}O_4SiNa$ requires $[M+Na]^+$ 309.1493; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 0.09 (6H, s, $Si(CH_3)_2$), 0.90 (9H, s, $SiC(CH_3)_3$), 1.75–1.86 (2H, m, 6-H_2), 2.51 (1H, dd, *J* 17.1 and 7.1 Hz, 4-H_aH_b), 2.60 (1H, dd, *J* 17.1 and 6.3 Hz, 4-H_aH_b), 3.76 (3H, s, OCH_3), 3.85 (1H, m, 7-H_aH_b), 3.93 (1H, m, 7-H_aH_b), 4.08 (1H, m,

5-H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ −5.64, −5.60, 18.1, 25.8, 27.2, 37.1, 52.6, 62.2, 69.9, 74.4, 86.2, 154.0; ν_{max} 3407, 2931, 2239, 1713, 1436, 1251, 1073, 835 cm^{-1}; *m*/*z* (ESI$^+$) 287.2 [M+H]$^+$.

3.4. (*R*)-6-(2-(*tert*-Butyldimethylsilyloxy)ethyl)-5,6-dihydro-4-methoxy-2*H*-pyran-2-one 6

To a solution of the acetylene **10** (0.96 g, 3.35 mmol) in methanol (20 mL) at 0 °C was added sodium methoxide (0.70 M in methanol, 1.44 mL) dropwise. The mixture was stirred at 0 °C for 1 h and then at ambient temperature for 16 h. The mixture was diluted with saturated ammonium chloride and extracted with ethyl acetate (4×10 mL), dried ($MgSO_4$) and concentrated in vacuo. Flash column chromatography (ethyl acetate/petrol 1:2) afforded the title compound **6** (765 mg, 80%) as a colourless oil. $[\alpha]_D^{26}$ −46.7 (*c* 2.00, $CHCl_3$). Found: [M+Na]$^+$ 309.1493. $C_{14}H_{26}O_4SiNa$ requires [M+Na]$^+$ 309.1493; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 0.04 (6H, s, $Si(CH_3)_2$), 0.87 (9H, s, $SiC(CH_3)_3$), 1.83 (1H, m, 6-C*H*$_a$H$_b$), 1.97 (1H, m, 6-CH$_a$*H*$_b$), 2.37 (1H, dd, *J* 17.1 and 3.7 Hz, 5-H$_{eq}$), 2.53 (1H, dd, *J* 17.1, 12.0 and 1.4 Hz, 5-H$_{ax}$), 3.73 (3H, s, OCH_3), 3.76 (1H, m, C*H*$_a$H$_b$OSi), 3.81 (1H, m, CH$_a$*H*$_b$OSi), 4.56 (1H, m, 6-H), 5.13 (1H, d, 1.4 Hz, 3-H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ −5.5, −5.4, 18.2, 25.8, 33.2, 37.6, 56.0, 58.5, 73.1, 90.3, 167.3, 172.9; ν_{max} 2929, 1710, 1625, 1251, 1220, 1090, 840, 776 cm^{-1}; *m*/*z* (ESI$^+$) 287.2 [M+H]$^+$.

3.5. (*S*)-3,4-Dihydro-10-hydroxy-3-(2-(*tert*-butyldimethylsilyloxy)ethyl)-7,9-dimethoxy-1*H*-naphtho[2,3-*c*]pyran-1-one 11

To a solution of diisopropylamine (0.60 mL, 4.28 mmol) in THF (15 mL) at −60 °C was added *n*-butyllithium (1.6 M in hexane, 2.7 mL, 4.28 mmol). The reaction mixture was allowed to warm to −10 °C and stirred for 15 min. After cooling to −70 °C, methyl 2,4-dimethoxy-6-methylbenzoate **5** (450 mg, 2.14 mmol) in THF (2 mL) was added and stirring was continued for 10 min at −70 °C. To the resultant red solution was added the (*R*)-lactone **6** (612 mg, 2.14 mmol) in THF (2 mL) and the mixture was allowed to warm to 0 °C over 30 min. The reaction was quenched with saturated ammonium chloride (10 mL), extracted with ethyl acetate (3×10 mL) and the combined organic layers were washed with brine, dried ($MgSO_4$) and concentrated in vacuo. Repeated flash column chromatography (ethyl acetate/petrol 1:4 then dichloromethane/ethyl acetate 19:1) afforded the title compound **11** (330 mg, 36%) as an oil. $[\alpha]_D^{22}$ +10.2 (*c* 0.16, CH_2Cl_2). Found: [M+Na]$^+$ 455.1862. $C_{23}H_{32}O_6SiNa$ requires [M+Na]$^+$ 455.1860; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 0.07 (6H, s, $Si(CH_3)_2$), 0.89 (9H, s, $SiC(CH_3)_3$), 1.94 (1H, m, 3-C*H*$_a$H$_b$), 2.07 (1H, m, 3-CH$_a$*H*$_b$), 3.01 (2H, m, 4-H$_2$), 3.82 (1H, m, C*H*$_a$H$_b$OSi), 3.88 (1H, m, CH$_a$*H*$_b$OSi), 3.91 (3H, s, OCH_3), 3.99 (3H, s, OCH_3), 4.75 (1H, m, 3-H), 6.47 (1H, d, *J* 2.2 Hz, ArH), 6.58 (1H, d, *J* 2.2 Hz, ArH), 6.85 (1H, s, ArH), 13.14 (1H, s, 10-OH); ^{13}C NMR ($CDCl_3$, 125 MHz) δ −5.42, −5.40, 18.2, 25.9, 33.6, 37.8, 55.4, 56.2, 58.6, 76.5, 98.3, 98.9, 100.9, 110.7, 115.3, 134.5, 141.5, 160.6, 161.7, 164.0, 171.0; ν_{max} 3401, 2948, 1719, 1632, 1608, 1583, 1375, 1254, 1208, 1164, 1110, 1050, 829 cm^{-1}; *m*/*z* (ESI$^-$) 317.1 [M−TBS]$^-$.

3.6. (*S*)-3,4-Dihydro-10-hydroxy-3-(2-hydroxyethyl)-7,9-dimethoxy-1*H*-naphtho[2,3-*c*]pyran-1-one 12

To the silyl ether **11** (1.54 g, 3.66 mmol) in THF (60 mL) was added 10% hydrochloric acid (40 mL). After stirring overnight, the mixture was concentrated in vacuo. The residue was diluted with H_2O (10 mL) and extracted with ethyl acetate (3×15 mL). The combined organic layers were washed with H_2O (10 mL), dried ($MgSO_4$) and concentrated in vacuo. The crude solid was recrystallized from acetone to afford **12** (0.490 g, 70%) as colourless needles, mp 163–164 °C. $[\alpha]_D^{19}$ +7.16 (*c* 0.25, acetone). Found: C 64.54, H 5.60%. Anal. Calcd for $C_{17}H_{18}O_6$: C 64.14, H 5.70. Found: [M+H]$^+$ 319.1176. $C_{17}H_{19}O_6$ requires [M+H]$^+$ 319.1176; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 2.01 (1H, m, 3-C*H*$_a$H$_b$), 2.12 (1H, m, 3-CH$_a$*H*$_b$), 3.02 (2H, m, 4-H$_2$), 3.90 (1H, m, C*H*$_a$H$_b$OH), 3.91 (3H, s, OCH_3), 3.96 (1H, m, CH$_a$*H*$_b$OH), 3.99 (3H, s, OCH_3), 4.80 (1H, m, 3-H), 6.47 (1H, d, *J* 2.4 Hz, ArH), 6.58 (1H, d, *J* 2.4 Hz, ArH), 6.85 (1H, s, ArH), 13.07 (1H, s, 10-OH); ^{13}C NMR ($CDCl_3$, 125 Hz) δ 33.6, 37.4, 55.4, 56.2, 58.6, 76.8, 98.3, 98.9, 100.7, 110.6, 115.4, 134.2, 141.5, 160.6, 161.8, 164.1, 170.9; ν_{max} 3412, 2947, 1632, 1606, 1583, 1371, 1340, 1171, 1112, 787 cm^{-1}; *m*/*z* (ESI$^+$) 319.1 [M+H]$^+$.

3.7. (*S*)-3,4-Dihydro-10-hydroxy-7,9-dimethoxy-1*H*-naphtho[2,3-*c*]pyran-1-one-3-acetic acid methyl ester 13

To alcohol **12** (220 mg, 0.69 mmol) in dichloromethane (7 mL) were added [bis(acetoxy)iodo]benzene (267 mg, 0.83 mmol) and TEMPO (11 mg, 0.07 mmol) and the mixture was stirred at ambient temperature for 16 h. The reaction mixture was then diluted with aq sodium thiosulfate (10 mL) and extracted with chloroform (4×5 mL). The combined organic layers were dried ($MgSO_4$) and concentrated in vacuo to give an oily residue. The residue, containing the crude intermediate aldehyde, was dissolved in a mixture of acetone (3 mL), *tert*-butanol (5 mL) and water (2 mL). 2-Methyl-2-butene (1 mL) was added followed by sodium dihydrogenphosphate (323 mg) and sodium chlorite (125 mg, 1.38 mmol) and the mixture was stirred vigorously for 3 h. After addition of 2 M hydrochloric acid (2 mL) the mixture was extracted with chloroform (6×5 mL) and the combined organic layers were dried ($MgSO_4$) and concentrated in vacuo. The resultant crude acid was dissolved in methanol (10 mL) with concentrated sulfuric acid (0.05 mL) and the solution was stirred at ambient temperature for 16 h. After concentration of the mixture in vacuo, water (10 mL) was added and the mixture was extracted with chloroform (4×5 mL) and the combined organic layers were dried ($MgSO_4$) and concentrated in vacuo. Flash column chromatography (ethyl acetate/petrol 2:1) afforded the title compound **13** (200 mg, 84%, three steps from **12**) as a pale yellow oil. $[\alpha]_D^{18}$ −20.2 (*c* 1.14, $CHCl_3$). Found: [M+Na]$^+$ 369.0944. $C_{18}H_{18}O_7Na$ requires [M+Na]$^+$ 369.0945; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 2.74 (1H, dd, *J* 16.2 and 6.8 Hz, 3-C*H*$_a$H$_b$), 2.95 (1H, dd, *J* 16.2 and 6.4 Hz, 3-CH$_a$*H*$_b$), 3.02 (1H, dd, *J* 15.9 and 10.6 Hz, 4-H$_{ax}$), 3.10 (1H, dd, *J* 15.9 and 3.4 Hz, 4-H$_{eq}$), 3.74 (3H, s, OCH_3), 3.90 (3H, s, OCH_3), 3.98 (3H, s, OCH_3), 5.01 (1H, m, 3-H), 6.46 (1H, d, *J* 2.2 Hz, ArH), 6.56 (1H, d, *J* 2.2 Hz, ArH), 6.85 (1H, s, ArH), 12.96 (1H, s, 10-OH); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 33.0, 39.4, 52.1, 55.5, 56.2, 75.0, 98.5, 99.0, 100.5, 110.7, 115.6, 133.4, 141.5, 160.7, 161.9, 164.2, 170.0, 170.3; ν_{max} 2931, 1740, 1634, 1609, 1375, 1209, 1165, 1112, 750 cm^{-1}; *m*/*z* (ESI$^+$) 347.1 [M+H]$^+$.

3.8. (*S*)-3,4-Dihydro-9,10-dihydroxy-7-methoxy-1*H*-naphtho[2,3-*c*]pyran-1-one-3-acetic acid methyl ester [(*S*)-semiviriditoxin] 2

To ester **13** (24 mg, 0.069 mmol) in dichloromethane (2 mL) at 0 °C was added boron trichloride (0.55 mL, 0.55 mmol). The mixture was allowed to warm to ambient temperature and stirred for a further 6 h. Water (3 mL) was added and the mixture was stirred vigorously for 30 min before being extracted with dichloromethane (3×5 mL). The combined organic layers were dried ($MgSO_4$) and concentrated in vacuo. Purification by flash column chromatography (chloroform/ethyl acetate/formic acid, 50:1:0.1) afforded unreacted methyl ether **13** (6 mg, 25%) and (*S*)-semiviriditoxin **2** (13 mg, 57%; 75% based on recovered **13**) as colourless crystals, mp 169–171 °C (lit.[5] mp 168–170 °C). $[\alpha]_D^{23}$ −8.60 (*c* 0.06, $CHCl_3$) {lit.[5] $[\alpha]_D^{20}$ −9.0 (*c* 0.21, $CHCl_3$)}; CD (trifluoroethanol) 217 ($\Delta\varepsilon$ −6.2), 237 (+0.5), 262 (−4.8), 320 (+0.5), 370 nm (+1.2). Found: [M+Na]$^+$ 355.0788. $C_{17}H_{16}O_7Na$ requires [M+Na]$^+$ 355.0788; ^{1}H NMR ($CDCl_3$,

500 MHz) δ 2.78 (1H, dd, *J* 16.4 and 6.6 Hz, 3-CH_aH_b), 2.98 (1H, dd, *J* 16.4 and 6.6 Hz, 3-CH$_a$$H_b$), 3.05 (1H, ddd, *J* 16.0, 10.8 and 1.4 Hz, 4-H_{ax}), 3.14 (1H, ddd, *J* 16.0, 3.4 and 0.8 Hz, 4-H_{eq}), 3.76 (3H, s, CO_2CH_3), 3.89 (3H, s, 7-OCH_3), 5.06 (1H, m, 3-H), 6.55 (1H, d, *J* 2.2 Hz, 8-H), 6.59 (1H, d, *J* 2.2 Hz, 6-H), 6.91 (1H, br s, 5-H), 9.44 (1H, s, 9-OH), 13.59 (1H, s, 10-OH); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 32.6, 39.4, 52.2, 55.5, 75.8, 99.0, 99.6, 101.7, 108.4, 116.3, 132.2, 140.6, 158.6, 162.8, 163.1, 169.8, 170.7; ν_{max} 3329, 2930, 2852, 1738, 1649, 1627, 1582, 1160, 750 cm^{-1}; *m*/*z* (ESI$^+$) 333.1 [M+H]$^+$. The ^{1}H and ^{13}C NMR data were in agreement with the literature.[5]

3.9. Ethyl (*R*)-5-hydroxyhex-2-ynoate 15

To a solution of ethyl propiolate (0.63 g, 6.42 mmol) in THF (10 mL) at −90 °C was added *n*-butyllithium (1.6 M in hexane, 4.0 mL, 6.42 mmol) and the mixture was stirred for 30 min at −90 °C. Boron trifluoride/diethyl etherate (0.82 mL, 6.42 mmol) was added followed by a solution of (*R*)-propylene oxide **14** (0.25 g, 4.30 mmol) in THF (2 mL) and stirring was continued for 2 h at −90 °C. Saturated ammonium chloride was added and the mixture was extracted with diethyl ether (3×5 mL) and the combined organic layers washed with brine, dried ($MgSO_4$) and concentrated in vacuo. Flash column chromatography (ethyl acetate/petrol 1:3) afforded the title compound **15** (0.55 g, 82%) as a colourless oil. $[\alpha]_D^{24}$ −9.85 (*c* 3.05, $CHCl_3$). Found: [M+Na]$^+$ 179.0680. $C_8H_{12}O_3Na$ requires [M+Na]$^+$ 179.0679; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 1.30 (3H, d, *J* 6.4 Hz, CH_3), 1.31 (3H, t, *J* 7.1 Hz, CH_2CH_3), 1.99 (1H, br s, OH), 2.49 (1H, dd, *J* 17.1 and 6.3 Hz, 4-H_aH_b), 2.53 (1H, dd, *J* 17.1 and 5.3 Hz, 4-H_aH_b), 4.05 (1H, m, 5-H), 4.22 (2H, q, *J* 7.1 Hz, CH_2CH_3); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 14.0, 22.6, 29.1, 62.0, 65.9, 75.1, 85.5, 153.6; ν_{max} 3408, 2978, 2234, 1705, 1251, 1067, 752 cm^{-1}; *m*/*z* (ESI$^+$) 156.9 [M+H]$^+$.

3.10. (*R*)-5,6-Dihydro-4-methoxy-6-methyl-2*H*-pyran-2-one 16

To a solution of the acetylene **15** (300 mg, 1.92 mmol) in methanol (10 mL) at 0 °C was added sodium methoxide (0.72 M in methanol, 0.65 mL) dropwise. The mixture was stirred at 0 °C for 1 h and then at ambient temperature for 16 h. The mixture was diluted with water (15 mL), 2 M hydrochloric acid (1 mL) and extracted with chloroform (3×5 mL), dried ($MgSO_4$) and concentrated in vacuo. Flash column chromatography (ethyl acetate/petrol 2:1) afforded the title compound **16** (218 mg, 80%) as a colourless solid, mp 58–59 °C (lit.[9] mp 61 °C). $[\alpha]_D^{24}$ −170 (*c* 1.00, $CHCl_3$) {lit.[9] $[\alpha]_D^{25}$ −168.09 (*c* 1.2, $CHCl_3$)}. Found: [M+Na]$^+$ 165.0523. $C_7H_{10}O_3Na$ requires [M+Na]$^+$ 165.0522; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 1.40 (3H, d, *J* 6.3 Hz, 6-CH_3), 2.31 (1H, dd, *J* 17.1 and 3.9 Hz, 5-H_{eq}), 2.42 (1H, ddd, *J* 17.1, 11.7 and 1.5 Hz, 5-H_{ax}), 3.71 (3H, s, OCH_3), 4.49 (1H, m, 6-H), 5.10 (1H, d, *J* 1.5 Hz, 3-H); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 20.4, 34.5, 55.9, 72.1, 90.1, 167.3, 172.7; ν_{max} 2982, 1692, 1620, 1393, 1240, 1205, 1053, 1005, 990, 851 cm^{-1}; *m*/*z* (ESI$^+$) 142.9 [M+H]$^+$. The ^{1}H and ^{13}C NMR data were in agreement with the literature.[9]

3.11. (*R*)-3,4-Dihydro-10-hydroxy-7-methoxy-9-methoxyethoxymethoxy-3-methyl-1*H*-naphtho[2,3-*c*]pyran-1-one 18

To a solution of diisopropylamine (0.39 mL, 2.74 mmol) in THF (12 mL) at −60 °C was added *n*-butyllithium (1.6 M in hexane, 1.7 mL, 2.74 mmol). The reaction mixture was allowed to warm to −10 °C and stirred for 15 min. After cooling to −70 °C, *ortho*-toluate **17** (320 mg, 1.12 mmol) in THF (1 mL) was added and stirring was continued for 10 min at −70 °C. To the resultant red solution was added the (*R*)-lactone **16** (160 mg, 1.12 mmol) in THF (1 mL) and the mixture was allowed to warm to 0 °C over 30 min. The reaction was quenched with saturated ammonium chloride (10 mL), extracted with ethyl acetate (3×10 mL) and the combined organic layers were washed with brine, dried ($MgSO_4$) and concentrated in vacuo. Flash column chromatography (ethyl acetate/petrol 1:2) afforded the title compound **18** (160 mg, 39%) as colourless needles, mp 93–94 °C. $[\alpha]_D^{21}$ −3.45 (*c* 0.82, $CHCl_3$). Found: [M+Na]$^+$ 385.1258. $C_{19}H_{22}O_7Na$ requires [M+Na]$^+$ 385.1258; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 1.53 (3H, d, *J* 6.3, 3-CH_3), 2.97 (2H, m, 4-H_2), 3.39 (3H, s, OCH_3), 3.61 (2H, m, CH_2), 3.89 (3H, s, 7-OCH_3), 3.96 (2H, m, CH_2), 4.71 (1H, m, 3-H), 5.43 (2H, s, OCH_2O), 6.65 (1H, d, *J* 2.3 Hz, ArH), 6.77 (1H, d, *J* 2.3 Hz, ArH), 6.85 (1H, s, ArH), 13.05 (1H, s, 10-OH); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 20.6, 34.9, 55.3, 58.9, 68.1, 71.5, 75.6, 94.5, 100.6, 100.7, 103.2, 111.2, 115.2, 134.0, 141.2, 157.5, 161.4, 163.9, 171.1; ν_{max} 2919, 1652, 1610, 1582, 1387, 1261, 1168, 1149, 1125, 1092, 1049, 1023, 938, 845 cm^{-1}; *m*/*z* (ESI$^+$) 363.2 [M+H]$^+$.

3.12. (*R*)-3,4-Dihydro-9,10-dihydroxy-7-methoxy-3-methyl-1*H*-naphtho[2,3-*c*]pyran-1-one [(*R*)-semivioxanthin] 3

To the methoxyethoxymethyl ether **18** (16 mg, 0.044 mmol) in THF (1 mL) was added concentrated hydrochloric acid (0.05 mL) and the solution was stirred at ambient temperature for 8 h. After diluting with chloroform (4 mL) and ethyl acetate (2 mL) and stirring for 15 h, water (5 mL) was added and the mixture was extracted with chloroform (3×5 mL) and the combined organic layers dried ($MgSO_4$) and concentrated in vacuo. Flash column chromatography (chloroform/ethyl acetate/formic acid 95:4:1) afforded (*R*)-semivioxanthin **3** (8 mg, 66%) as a pale yellow solid, mp 174–177 °C (lit.[18] mp 185 °C), CD (trifluoroethanol) 216 ($\Delta\varepsilon$ −5.0), 238 (+0.8), 266 (−3.1), 323 (+0.4), 363 nm (+1.0). Found: [M−H]$^-$ 273.0776. $C_{15}H_{13}O_5$ requires [M−H]$^-$ 273.0769; ^{1}H NMR ($CDCl_3$, 500 MHz) δ 1.55 (3H, d, *J* 6.3, 3-CH_3), 2.96 (1H, ddd, *J* 16.0, 10.3 and 1.4 Hz, 4-H_{ax}), 3.02 (1H, ddd, *J* 16.0, 3.8 and 0.9 Hz, 4-H_{eq}), 3.89 (3H, s, 7-OCH_3), 4.76 (1H, m, 3-H), 6.54 (1H, d, *J* 2.4 Hz, ArH), 6.57 (1H, d, *J* 2.4 Hz, ArH), 6.88 (1H, br s, ArH), 9.48 (1H, s, 9-OH), 13.77 (1H, s, 10-OH); ^{13}C NMR ($CDCl_3$, 125 MHz) δ 20.8, 34.7, 55.4, 76.5, 99.2, 99.5, 101.5, 108.3, 116.0, 133.1, 140.5, 158.6, 162.7, 163.0, 171.5; ν_{max} 3383, 2930, 1637, 1580, 1381, 1323, 1246, 1158, 1121, 1083, 1033, 841 cm^{-1}; *m*/*z* (ESI$^-$) 273.0 [M−H]$^-$. The ^{1}H and ^{13}C NMR data were in agreement with the literature.[18]

Acknowledgements

Financial support from the Australian Research Council through the Centres of Excellence program is gratefully acknowledged. Professor Carl Schiesser, The University of Melbourne, is acknowledged for useful discussions.

References and notes

1. Vollmer, W. *Appl. Microbiol. Biotechnol.* **2006**, *73*, 37–47.
2. Wang, J.; Galgoci, A.; Kodali, S.; Herath, K. B.; Jayasuriya, H.; Dorso, K.; Vicente, F.; Gonzalez, A.; Cully, D.; Bramhill, D.; Singh, S. *J. Biol. Chem.* **2003**, *278*, 44424–44428.
3. Weisleder, D.; Lillehoj, E. B. *Tetrahedron Lett.* **1971**, *12*, 4705–4706.
4. Suzuki, K.; Nozawa, K.; Nakajima, S.; Kawai, K. *Chem. Pharm. Bull.* **1990**, *38*, 3180–3181.
5. Ayer, W. A.; Craw, P. A.; Nozawa, K. *Can. J. Chem.* **1991**, *69*, 189–191.
6. Arnone, A.; Assante, G.; Montorsi, M.; Nasini, G. *Phytochemistry* **1995**, *38*, 595–597.
7. (a) Evans, G. E.; Leeper, F. J.; Murphy, J. A.; Staunton, J. *J. Chem. Soc., Chem. Commun.* **1979**, 205–206; (b) Carpenter, T. A.; Evans, G. E.; Leeper, F. J.; Staunton, J.; Wilkinson, M. R. *J. Chem. Soc., Perkin Trans. 1* **1984**, 1043–1051.
8. Deshpande, V. H.; Khan, R. A.; Rai, B.; Ayyangar, N. R. *Synth. Commun.* **1993**, *23*, 2337–2345.
9. Drochner, D.; Müller, M. *Eur. J. Org. Chem.* **2001**, 211–215.
10. Donner, C. D.; Gill, M.; Tewierik, L. M. *Molecules* **2004**, *9*, 498–512.
11. Donner, C. D. *Tetrahedron Lett.* **2007**, *48*, 8888–8890.
12. Volkmann, R. A.; Kelbaugh, P. R.; Nason, D. M.; Jasys, V. J. *J. Org. Chem.* **1992**, *57*, 4352–4361.
13. Mori, K.; Ikunaka, M. *Tetrahedron* **1984**, *40*, 3471–3479.

14. Yamaguchi, M.; Hirao, I. *Tetrahedron Lett.* **1983**, *24*, 391–394.
15. (a) Carlson, R. M.; Oyler, A. R. *Tetrahedron Lett.* **1974**, *15*, 2615–2618; (b) Carlson, R. M.; Oyler, A. R.; Peterson, J. R. *J. Org. Chem.* **1975**, *40*, 1610–1616.
16. The addition of alcohols to α,β-acetylenic esters is known to lead predominantly to the *Z*-isomer: Winterfeldt, E. *Angew. Chem., Int. Ed. Engl.* **1967**, *6*, 423–434.
17. Tan, N. P. H.; Donner, C. D. *Tetrahedron Lett.* **2008**, *49*, 4160–4162.
18. Zeeck, A.; Ruβ, P.; Laatsch, H.; Loeffler, W.; Wehrle, H.; Zähner, H.; Holst, H. *Chem. Ber.* **1979**, *112*, 957–978.
19. Although **2** and **3** possess different descriptors, (*S*) and (*R*), respectively, they are stereochemically analogous and would therefore be expected to exhibit similar Cotton effects in their circular dichroism spectra.