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Total Synthesis of the Nonadjacently Linked Bis-tetrahydrofuran Acetogenin Bullatanocin (Squamostatin C)

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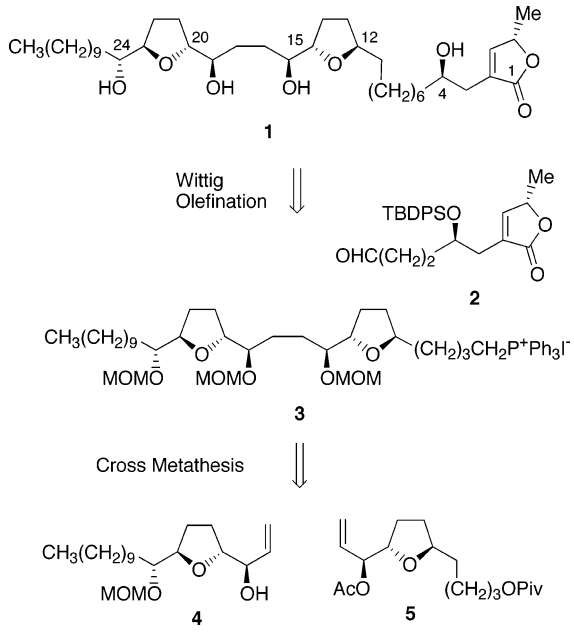
The total synthesis of the nonadjacently linked bis-THF acetogenin bullatanocin (squamostatin C) is described. The synthetic strategy is a modular one based on three components, two mono-THF alkenes and a butenolide precursor, and the olefin cross-metathesis and Wittig olefination as the segment-coupling reactions. The synthesis confirms the structure of the natural product, and its convergent design makes it especially attractive for analogue synthesis.

Introduction

The potent antitumor activity of the tetrayhdrofuran-containing acetogenins has contributed to considerable synthetic activity in this area.[1] Three major subtypes, depending on the number and connectivity of the THF rings, have been identified: mono-THFs, adjacently linked bis-THFs, and nonadjacently linked bis-THFs. Several syntheses in the first two groups have been reported.[2] The nonadjacently linked bis-THF structures have not been as well explored. Three naturally occurring structures have been prepared by total synthesis.[3] Four total syntheses of mucocin an unusual, structurally related, TH−-THP analogue, are also noteworthy.[4]

Bullatanocin **1** (squamostatin C) is typical of the nonadjacently linked bis-THF acetogenins, both in terms of biological activity and structure (Scheme 1). Bullatanocin isolated from the bark of *Annona bullata* Rich (Annonaceae) by the Mclaughlin group exhibits physical data identical to that of squamostatin C found in the seeds of *Annona squamosa*.[5] The material isolated by Mclaughlin shows ED_{50} of less than 10^{-8} μg/mL against the colon cell line HT-29 and the lung cell line A-549, which is over 10 000 times the activity of adriamycin. The structure of bullatanocin comprises two 2,5-trans-disubstituted THFs bridged by a 1,4-dihydroxybutyl linker. Structures containing two trans THFs account for the largest subgroup. There are smaller families with one trans and one cis-THF or two cis THFs. The relative

SCHEME 1



(1) (a) Rupprecht, J. K.; Hui, Y.-H.; McLaughlin, J. L. *J. Nat. Prod.* **1990**, *53*, 237−278. (b) Fang, X.; Rieser, M. J.; Gu, Z.; Zhao, G.; McLaughlin, J. L. *Phytochem. Anal.* **1993**, *4*, 27−48. (c) Zeng, L.; Ye, Q.; Oberlis, N. H.; Shi, G.; Gu, Z.-M.; He, K.; McLaughlin, J. L. *Nat. Prod. Rep.* **1996**, *13*, 275−306. (d) Alali, F. Q.; Liu, X.-X.; McLaughlin, J. L. *J. Nat. Prod.* **1999**, *62*, 504−540.

(2) Recent reviews on synthesis of THF acetogenins: (a) Zafra-Polo, M. C.; Figadere, B.; Gallardo, T.; Tormo, J. R. Cortes, D.; Figadère, B. *Phytochemistry* **1998**, *48*, 1087−1117. (b) Figadère, B. *Acc. Chem. Res.* **1995**, *28*, 359−365. (c) Koert, U. *Synthesis* **1995**, 115−132. (d) Hoppe, R.; Scharf, H.-D. *Synthesis* **1995**, 1447−1464. (e) Casiraghi, G.; Zanardi, F.; Battistini, L.; Rassu, G.; Appendino, G.*Chemtracts-Org. Chem.* **1998**, *11*, 803−827.

(3) (a) Marshall J. A.; Jiang, H. *J. Org. Chem.* **1998**, *63*, 7066−7071. (b) Szlosek, M.; Peyrat, J.-F.; Chaboche, C.; Franck, X.; Hocquemiller, R.; Figadere, B. For synthesis of an advanced precursor to an nonadjacent bis-THF acetogenin, see: (c) *New J. Chem.* **2000**, *24*, 337−342. Makabe, H.; Tanaka, A.; Oritan, T. *Tetrahedron* **1998**, *54*, 6329−6340.

(4) (a) Neogi, P.; Doundoulakis, T.; Yazbak, A.; Sinha, S. C.; Sinha, S. C.; Keinan, E. *J. Am. Chem. Soc.* **1998**, *120*, 11279−11284. (b) Hoppen, S.; Bäurle, S.; Koert, U. *Chem. Eur. J.* **2000**, *6*, 2382−2396. (c) Takahashi, S.; Kubota, A.; Nakata, T. *Angew. Chem., Int. Ed. Engl.* **2002**, *41*, 4751−4754. (d) Evans, P. A.; Cui, J.; Gharpure, S. J.; Polosukhin, A.; Zhang, H.-R. *J. Am. Chem. Soc.* **2003**, *125*, 14702−14703.

(5) (a) Bullatanocin: Gu, Z. G.; Fang, X.; Rieser, M. J.; Hui, Y.; Miesbauer, L. R.; Smith, D. L.; Wood, K. V.; McLaughlin, J. L. *Tetrahedron* **1993**, *49*, 745−754. Gu, Z.; Zeng, L.; Fang, X.; Colman-Saizarbitoria, T.; Huo, M.; McLaughlin, J. L. *J. Org. Chem.* **1994**, *59*, 5162−5172. (b) Squamostatin C: Shimada, H.; Nishioka, S.; Singh, S.; Sahai, M.; Fujimoto, Y. *Tetrahedron Lett.* **1994**, *35*, 3961−3964. (c) Fujimoto, Y.; Murasaki, C.; Shimada, H.; Nishioka, S.; Kakinuma, K.; Singh, S.; Singh, M.; Gupta, Y. K.; Sahai, M. *Chem. Pharm. Bull.* **1994**, *42*, 1175−1184.

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stereochemistry at the carbinol carbons is variable. As in the case of bullatanocin, stereochemistry is generally assigned through empirical rules formulated by NMR analysis of reference compounds. Although this approach has proven to be quite reliable, it still contains an element of uncertainty. Given the poor crystalline properties of these compounds, total synthesis remains an important tool for unambiguous structural determination.

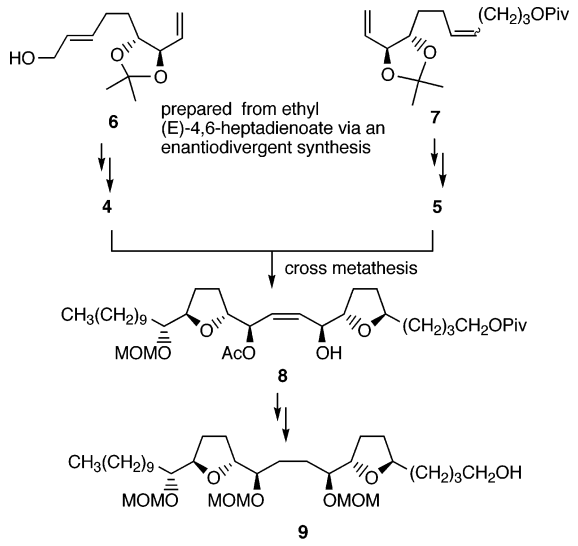
In view of the variety of natural analogues and the demand for unnatural structures for structure−activity investigations, modular syntheses that provide analogue libraries with different stereoisomeric THF subunits, hydrocarbon chain lengths, and butenolide substitutes are appealing (Scheme 1). We envisaged an approach to bullatanocin based on three components, butenolide **2** and THFs **4** and 6, and the Wittig olefination and olefin metathesis as the segment-coupling reactions. Thus, **1** could be accessed through a late-stage Wittig coupling of a bis-THF **3** and butenolide **2**,[6] and **2** via an olefin cross-metathesis[7] of THF alkenes **4** and **5**.[8] We have previously disclosed a successful plan for the cross-metathesis of the THF alkene subunits.[9] Herein,- is described the completion of the first total synthesis of bullatanocin (squamostatin C), which supports the structure originally assigned to the natural product and lays the groundwork for the application of this synthetic strategy to analogous structures.

To summarize our earlier work, bis-THF alkene **8** was obtained through the cross-metathesis of THF alkenes **4** and **5**. Attributes of this synthesis are the convergent design, the experimental simplicity of the metathesis coupling step, and the straightforward and stereoselective preparation of **4** and **5** (via the iodoetherification of isopropylidene alkenes **6** and **7**.[10] Routine functional group conversions on **8** provided bis-THF **9** (Scheme 2).[11]

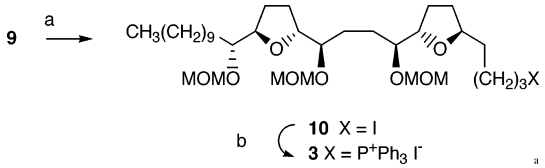
The first stage in the completion of the synthesis was the conversion of **9** to phosphonium salt **3** for the final Wittig coupling with aldehyde **2** (Scheme 3). Thus, treatment of the iodide derived from **9** with triphenylphosphine in the presence of Hunig's base provided **3**.

The known butenolide-aldehyde **2** was obtained by modification of the synthesis of Keinan and Sinha.[6] The original route involved preparation of methyl ester **15** from glutamic acid and transformation of **15** to **2** (Scheme 4). We found an alternative synthesis of **15** to be more practical. Thus, following the strategy employed by Hoye and co-workers,[12] the bis-pentenyl ether of hydroquinone, **11**, was subjected to double dihydroxylation with AD-mix-α. Three crystallizations of the crude product provided a mixture of *dl*-**12** and the meso isomer with a total R/S ratio that was estimated at 20/1 by NMR analysis of the tetra Mosher ester derivative (Supporting Information). Tetrol **12** was next transformed to primary alcohol **14** via a straightforward sequence of reactions. Standard processing of **14** furnished methyl ester **15**, which was converted to butenolide−aldehyde **2** via the identical

SCHEME 2

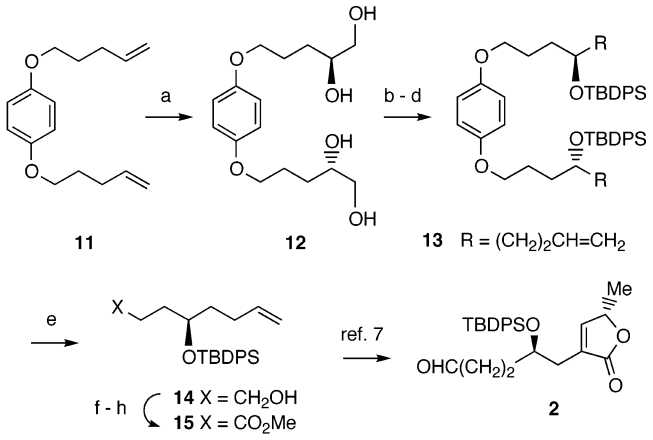


SCHEME 3[a]



[a] Reaction conditions: (a) Ph_3P, I_2, imidazole, PhH; (b) Ph_3P, $^iPr_2NEt$, CH_3CN.

SCHEME 4[a]



[a] Reaction conditions: (a) AD-mix-α; 50%; (b) (i) TsCl, py; (ii) K_2CO_3, MeOH, 84%; (c) allylMgBr, CuI, 72%; (d) TBDMSCl, imidazole, 99%; (e) CAN, CH_3CN-H_2O, 80%; (f) PCC, CH_2Cl_2; (g) $NaClO_2$, H_2O_2, 72%, two steps; (h) MeOH, DCC, DMAP, 88%.

(6) (a) Sinha, S. C.; Sinha, A.; Sinha, S. C.; Keinan, E. *J. Am. Chem. Soc.* **1997**, *119*, 12014−12015. (b) Avedissian, H.; Sinha, S. C.; Yazbak, A.; Sinha, A.; Neogi, P.; Sinha, S. C.; Keinan, E. *J. Org. Chem.* **2000**, *65*, 6035−6051.

(7) (a) Grubbs, R. H.; Chang, S. *Tetrahedron* **1998**, *42*, 2809. (b) Blackwell, H. E.; O'Leary, D. J.; Chatterjee, A. K.; Washenfelder, R. A.; Bussmann, D. A.; Grubbs, R. H. *J. Am. Chem. Soc.* **2000**, *122*, 58−71. (c) Chatterjee, A. K.; Choi, T.-L.; Sanders, D. P.; Grubbs, R. H. *J. Am. Chem. Soc.* **2003**, *125*, 11360−11370.

(8) For a conceptually similar strategy using a ring-closing metathesis approach, see ref 4d.

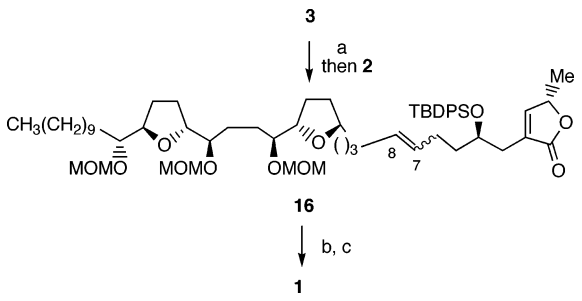
(9) For a preliminary account: Zhu, L.; Mootoo, D. R. *Org. Lett.* **2003**, *5*, 3475−3478.

(10) (a) Zhang, H.; Mootoo, D. R. *J. Org. Chem.* **1995**, *60*, 8134−8135. (b) Zhang, H.; Seepersaud, M.; Seepersaud, S.; Mootoo, D. R. *J. Org. Chem.* **1998**, *63*, 2049−2052. (c) Ruan, Z.; Mootoo, D. R. *Tetrahedron Lett.* **1999**, *40*, 49−52. (d) Dabideen, D.; Ruan, Z.; Mootoo, D. R. *Tetrahedron* **2002**, *58*, 2077−2084.

(11) For a detailed description of the synthesis of **9** and stereochemical analysis of **4** and **5**, see ref 8 and the Supporting Information.

(12) Hoye, T. R.; Mayer, M. J.; Vos, T. J.; Ye, Z. *J. Org. Chem.* **1998**, *63*, 8554−8557.

SCHEME 5[a]



[a] Reaction conditions: (a) BuLi, THF, −78 °C, 44%; (b) $Rh(PPh_3)_3Cl$, H_2, 89%; (c) AcCl, MeOH−CH_2Cl_2, 71%.

sequence used by Keinan and Sinha. The ^1H and ^{13}C NMR spectra for compound **2** and the material obtained by this group were essentially identical. Unfortunately, the unavailability of optical rotation data for the original sample of **2** did not allow for a direct comparison of optical purity.

Treatment of the ylide generated by treatment of phosphonium salt **3** with butyllithium, with 2 equivalents of aldehyde **2**, following the reported protocol,[6] afforded alkene **16** in 44% yield as an undetermined mixture of *E*/*Z* isomers (Scheme 5). Selective hydrogention of **16** and exposure of the product to methanolic HCl provided **1**. The ^1H and ^{13}C NMR were essentially identical to those for bullatanocin (squamostatin C, Supporting Information). The $[\alpha]_D$'s of **1** in $CHCl_3$ and MeOH were found to be 16.5 and 18.7 compared with corresponding values of 14.4 (bullatanocin) and 12.0 (squamostatin C), respectively.

In summary, the total synthesis of the acetogenin bullatanocin (squamostatin C) has been described, thereby supporting the structure originally assigned to the natural product. A feature of this synthesis is the three-component modular design. In principle, this approach should be applicable to stereoisomers and homologues of building blocks **2**, **4**, and **5** and is well suited to split synthesis of libraries of nonadjacently linked cyclic ether containing acetogenins.[13]

Experimental Section

Phosphonium Salt 3. A mixture of triphenylphosphine (139 mg, 0.13 mmol), imidazole (72 mg, 0.26 mmol), iodine (135 mg, 0.13 mmol), and benzene (100 mL) was stirred at rt for 30 min, at which time alcohol **9** (160 mg, 0.26 mmol) was introduced. The reaction mixture was stirred at reflux for 2 h, diluted with ethyl ether (100 mL), and filtered through a column of Florisil. The filtrate was concentrated under reduced pressure and the residue purified by FCC to afford iodide **10** (164 mg, 88%): R_f = 0.77 (50% EtOAc/petroleum ether); $[\alpha]^{22}_D$ +18.5 (c 2.92, $CHCl_3$); ^1H NMR ($CDCl_3$, 500 MHz) δ 4.80−4.82 (m, 3H), 4.63−4.67 (m, 3H), 3.95 (m, 3H), 3.85 (m, 1H), 3.38 (s, 3H), 3.37 (s, 6H), 3.1 (t, 2H, J = 7 Hz), 1.6−2.0 (m, 12H), 1.3−1.5 (m, 8H), 1.20 (s, br, 16H), 0.85 (t, 3H, J = 6.5 Hz); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 96.9, 81.7, 81.1, 80.1, 79.8, 79.1, 56.0, 34.9, 33.8, 32.5, 32.15, 31.5, 30.1, 29.9, 29.6, 28.8, 28.7, 27.6, 27.4, 25.8, 22.9, 14.4, 7.1; HRMS (ESI) calcd for $C_{33}H_{63}O_8INa$ (M + Na) 737.3465, found 737.3478.

Diisopropylethylamine (40 mL, 0.35 mmol) was added to a solution of **10** (163 mg, 0.23 mmol) and triphenylphosphine (240 mg, 0.92 mmol) in anhydrous toluene (4 mL) and acetonitrile (2 mL). The reaction mixture was heated at reflux under an atmosphere of argon for 24 h. Most of the solvent was then removed under reduced pressure. The resulting syrup was triturated with cold hexane. Drying of the residue under high vacuum for 12 h afforded **3** as a white solid (220 mg, 90%): R_f = 0.42 (80% EtOAc/20% MeOH); $[\alpha]^{22}_D$ +12 (*c* 1.6, $CHCl_3$); ^1H NMR ($CDCl_3$, 300 MHz) δ 7.6−7.9 (m, 15H), 4.5−4.9 (m, 6H), 3.80−4.00 (m, 3H), 3.60−3.80 (m, 1H), 3.40−3.50 (m, 3H), 3.36 (s, 3H), 3.35 (s, 3H), 3.34 (s, 3H), 1.30−2.00 (m, 20H), 1.20 (s, br, 18H), 0.85 (t, 3H, J = 6.5 Hz); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 135.7, 135.7, 134.4, 134.3, 131.3, 131.1, 97.4, 82.3, 81.7, 80.9, 80.7, 80.4, 79.5, 56.5, 35.7, 32.8, 32.6, 32.1, 30.6, 30.3, 30.1, 29.2, 29.2, 28.0, 27.7, 26.3, 23.4, 14.8; HRMS (ESI) calcd for $C_{51}H_{78}O_8P$ (M $-I^-$) 849.5434, found 849.5440.

Bis-THF-butenolide 16. BuLi (2.5 M solution in hexane, 12.3 mL, 0.029 mmol) was added dropwise to a solution of **3** (28 mg, 0.028 mmol) in dry THF (1 mL) at 0 °C, under an atmosphere of argon. The mixture was stirred for 30 min, and a solution of **2** (25 mg, 0.057 mmol) in dry THF (1 mL) was added dropwise at −78 °C. The reaction was warmed to rt, quenched with saturated aqueous NH_4Cl, and extracted with ether. The combined organic phase was dried (Na_2SO_4), filtered, and concentrated under reduced pressure. FCC of the crude product provided **16** (12.6 mg, 44%): R_f = 0.30 (20% EtOAc/petroleum ether); $[\alpha]^{22}_D$ +7.4 (*c* 0.60, $CHCl_3$); ^1H NMR ($CDCl_3$, 500 MHz) δ 7.65−7.67 (m, 4H), 7.35−7.43 (m, 6H), 6.91 (s, 1H), 5.08−5.12 (m, 1H), 5.22−5.25 (m, 1H), 4.82−4.89 (m, 4H), 4.65−4.68 (m, 3H), 4.03 (t, 1H, J = 5.5 Hz), 3.96−3.97 (m, 3H), 3.85 (m, 1H), 3.44 (m, 3H), 3.39 (s, 9H), 2.45 (t, 2H, J = 5.5 Hz), 1.91−1.99 (m, 8H), 1.38−1.74 (m, 16H), 1.31 (d, 3H, J = 7 Hz), 1.2 (s, br, 16H), 1.0 (s, 9H), 0.85 (t, 3H, J = 5.5 Hz); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 173.9, 151.3, 136.0, 134.2, 130.8, 130.2, 129.9, 129.2, 127.8, 97.0, 81.8, 81.2, 80.3, 80.0, 79.4, 71.8, 56.0, 36.7, 35.8, 32.5, 32.2, 32.1, 31.6, 30.1, 30.0, 29.9, 29.6, 28.9, 28.8, 28.8, 27.6, 27.5, 27.4, 26.7, 25.9, 23.2, 23.0, 19.7, 19.3, 14.4; HRMS (ESI) calcd for $C_{59}H_{94}O_{11}Si$ (M + Na^+) 1029.6463, found 1029.6437.

Bullatanocin 1. Chlorotris(triphenylphosphine)rhodium-(I) (1.7 mg, 1.8 mmol) was added to a degassed solution of **16** (12 mg, 12 μmol) in benzene−EtOH (1:1, 0.5 mL), and the mixture was stirred under an atmosphere of hydrogen for 12 h. The solvent was removed under reduced pressure, and the residue was purified by FCC to give the 7,8-dihydro derivative of **16** (10.8 mg, 89%): R_f = 0.56 (30% EtOAc/70% petroleum ether); $[\alpha]^{22}_D$ +9.5 (c 0.42, $CHCl_3$); ^1H NMR ($CDCl_3$, 500 MHz) δ 7.63−7.70 (m, 4H), 7.36−7.42 (m, 6H), 6.91 (s, 1H), 4.84−4.89 (m, 4H), 4.68 (m, 3H), 3.98 (m, 4H), 3.85 (m, 1H), 3.45 (m, 3H), 3.39 (s, 9H), 2.43 (s, 2H), 1.90−2.00 (m, 8H), 1.00−1.70 (m, 23H), 1.6 (s, br, 16H), 1.0 (s, 9H), 0.88 (t, 3H, J = 7.0 Hz); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 151.2, 136.0, 134.4, 130.9, 129.8, 128.5, 127.7, 97.0, 81.8, 81.1, 80.3, 80.3, 80.0, 79.6, 72.1, 56.0, 36.8, 36.2, 32.6, 32.2, 31.7, 30.7, 30.2, 29.9, 29.7, 29.6, 28.9, 28.8, 28.8, 27.6, 27.5, 27.4, 26.6, 25.9, 25.2, 23.0, 22.7, 19.7, 19.3, 14.4; HRMS (ESI) calcd for $C_{59}H_{96}O_{11}Si$ (M + Na^+) 1031.6620, found 1031.6587.

AcCl (5%) in MeOH (0.15 mL) was added at rt to a solution of the material obtained in the previous step (3.8 mg, 3.8 μmol) in CH_2Cl_2 (0.5 mL). The mixture was stirred at this temperature for 2 h, diluted with CH_2Cl_2, and washed with a saturated aqueous $NaHCO_3$. The organic layer was dried (Na_2SO_4), filtered, and concentrated under reduced pressure. FCC of the residue afforded **1** (1.7 mg, 71%) and a mixture of less polar fractions (1.8 mg) that appeared to be partially deprotected products. For **1**: R_f = 0.36 (EtOAc); mp 97−99 °C (EtOAc) [lit.[5c] mp 95−97 °C]; $[\alpha]^{22}_D$ +18.7 (*c* 0.23, MeOH), +16.5 (*c* 0.23, $CHCl_3$) [lit.[5c] $[\alpha]^{22}_D$ +12.0 (*c* 0.20, MeOH), lit.[5a] $[\alpha]^{22}_D$ +14.4 (*c* 0.55, $CHCl_3$)]; ^1H NMR ($CDCl_3$, 500 MHz) δ 7.18 (s, 1H), 5.06 (dd, 1H, J = 6.5 Hz), 3.78−3.88 (m, 5H),

(13) For a recent example in THF acetogenin synthesis, see: Zhang, Q.; Lu, H.; Richard, C.; Curran, D. P. *J. Am. Chem. Soc.* **2004**, *126*, 36−37.

3.39−3.49 (m, 3H), 2.73 (s, br, 1H), 2.51−2.55 (m, 2H), 2.37−2.42 (m, 2H), 2.3 (s, br, 1H), 1.95−2.02 (m, 4H), 1.25−1.80 (m, 40H), 1.42 (d, 3H, J = 6.5 Hz), 0.88 (t, 3H, J = 7.0 Hz); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 174.6, 151.9, 131.2, 82.7, 82.0, 79.3, 78.0, 74.4, 74.3, 74.1, 70.0, 37.4, 35.6, 33.4, 32.4, 31.9, 29.9, 29.7, 29.6, 29.5, 29.4, 29.3, 28.7, 28.4, 26.1, 25.6, 25.5, 22.7, 19.1, 14.1; HRMS (ESI) calcd for $C_{37}H_{66}O_8$ ($M + Na^+$) 661.4655, found 661.4644.

Acknowledgment. We thank Professor Yoshinori Fujimoto for providing original ^{1}H and ^{13}C NMR charts for squamostatin C. Support was provided by NIGMS - SCORE grant 5 SO6 GM60654 to Hunter College. "Research Centers in Minority Institutions" award RR-03037 from the National Center for Research Resources of the NIH, which supports the infrastructure (and instrumentation) of the Chemistry Department at Hunter, is also acknowledged.

Note Added after ASAP Posting. Compound **9** in Scheme 2 was incorrectly labeled as compound **8** in the version posted ASAP April 7, 2004; the corrected version was posted April 12, 2004.

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

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