Hexapeptide Total Synthesis

Total Synthesis of Microsclerodermin E**

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Lithistid sponges are well-known among marine organisms for their ability to produce a diverse array of biologically active metabolites.[1] From several species of the lithistid sponge *Microscleroderma sp.* and *Theonella sp.*, Faulkner and co-workers have isolated a series of cyclic peptides named microsclerodermins A–I.[2] Structurally, each of them was shown to display several unique features, including a 23-membered cyclic hexapeptide core, featuring a very complex β-amino acid and three other unnatural amino acid residues. Preliminary studies have indicated that all these compounds possess intriguing antifungal activities and some of them show potent antitumor properties,[2,3] but further evaluation of their biological activities has been hampered by their relative scarcity. The biological activities and the structural complexity exhibited by the microsclerodermins have attracted much attention within the community of synthetic chemists. However, although some synthetic efforts towards these compounds have been reported,[4] no member of this family has been synthesized yet. Herein we wish to describe our total synthesis of microsclerodermin E. Notable elements of the synthesis are the concise assembly of its β-amino acid fragment, the assembly of the pyrrolidinone fragment with little racemization, and the effective macrocyclization at the Gly-GABOB site.

The retrosynthetic blueprint that guided this new campaign is defined in Figure 1. The macrocyclic core of the target molecule is disconnected into two fragments, the tetrapeptide

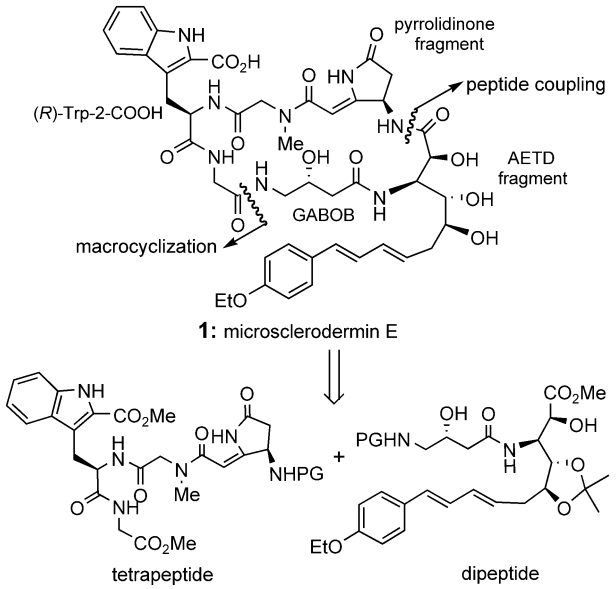

Figure 1. Retrosynthetic analysis of microsclerodermin E. PG = protecting group.

portion and the dipeptide unit. Further disconnections of these two parts reveals that we need to employ Gly and *N*-MeGly as the starting materials and develop some efficient methods for assembling four unusual amino acid units, which include 3-amino-10-(*p*-ethoxyphenyl)-2,4,5-trihydroxydeca-7,9-dienoic acid (AETD), 4-amino-3-hydroxybutanoic acid (GABOB), (*R*)-Trp-2-carboxylic acid, and a pyrrolidinone fragment.

Our approach towards the AETD fragment is shown in Scheme 1 (see scheme legends for abbreviations). Treatment of lithiated methyl phenyl sulfone in THF at −78 °C with oxirane **2** (derived from δ-gluconolactone as shown) in the presence of $BF_3{\cdot}Et_2O$[5] afforded the desired γ-hydroxy

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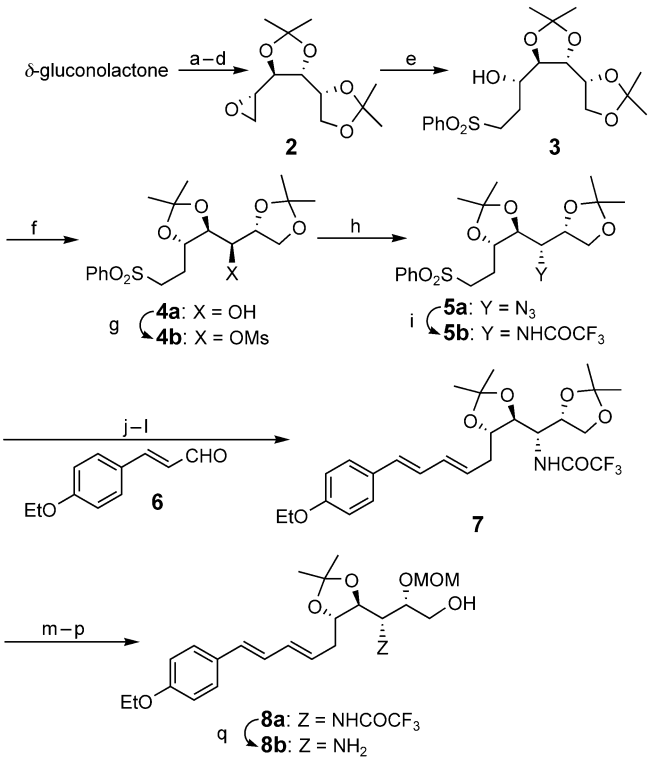
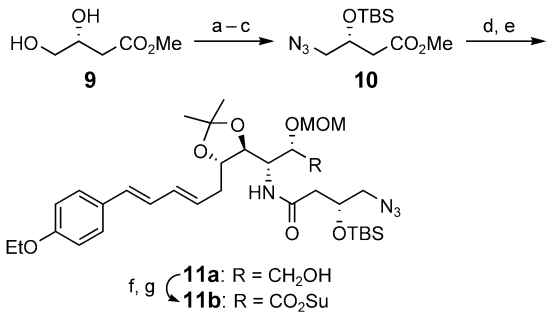


Scheme 1. Reagents and conditions: a) 2,2-dimethoxypropane, acetone, TsOH, 80%; b) $NaBH_4$/MeOH, 100%; c) TsCl, Py, 85%; d) K_2CO_3, MeOH, 89%; e) $PhSO_2CH_2Li$, $BF_3{\cdot}Et_2O$, −78°C, 84%; f) 1N HCl, THF then 2,2-dimethoxypropane, acetone, TsOH, 67%; g) MsCl, DMAP, Et_3N, 97%; h) NaN_3, DMF, 120°C, 83%; i) Pd/C, H_2 then $(CF_3CO)_2O$, Py, 62%; j) *n*BuLi, THF, −78°C then **6**; k) PhCOCl, DMAP, Et_3N; l) Na(Hg), MeOH, 77% for 3 steps; m) PPTS, MeOH, 45°C, 87%; n) TBSCl, imidazole, 92%; o) MOMCl, iPr_2NEt, 100%; p) TBAF, THF, 100%; q) $NaBH_4$, EtOH, 100%. DMAP = 4-dimethylaminopyridine, DMF = dimethylformamide, MOM = methoxymethyl, Ms = methanesulfonyl, PPTS = pyridinium *p*-toluenesulfonate, Py = pyridine, TBAF = tetrabutylammonium fluoride, TBS = *tert*-butyldimethylsilyl, Ts = toluenesulfonyl.

sulfone **3**. Subsequent exposure of **3** to 1N HCl and regioselective reprotection of the resultant alcohol with 2,2-dimethoxypropane produced **4a** in 67% yield. This approach relied on the preferential formation of the acetonides with less steric hindrance. Mesylation of the hydroxy function of **4a**, followed by azidation of the resultant mesylate **4b** furnished azide **5a**, which was hydrogenated over Pd/C, and then treated with trifluoroacetic anhydride to provide amide **5b**. Julia reaction[6] of **5b** with α,β-unsaturated aldehyde **6** afforded the desired diene **7**, together with its *E*/*Z* isomer in a ratio of 5:1. After the terminal acetonide in **7** was selectively removed with the assistance of PPTS,[7] the two liberated hydroxy groups were protected selectively with TBSCl and MOMCl, and the TBS ether was then cleaved with TBAF to deliver amide **8a**. Removal of the CF_3CO group in **8a** by reaction with $NaBH_4$[8] led to the free amine **8b**, which was required for the coupling with the GABOB unit.

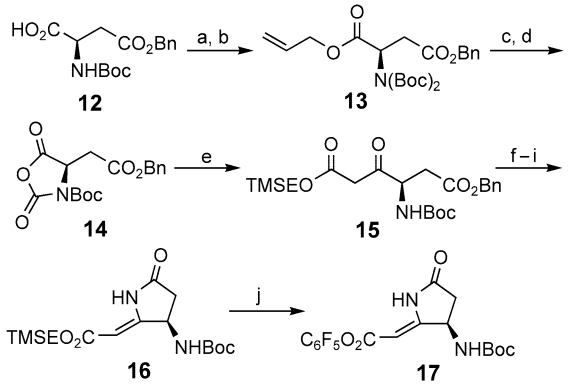
The GABOB unit **10** was constructed from methyl (*R*)-3,4-dihydroxybutanoate **9**[9] (Scheme 2). Selective tosylation[10] of the primary hydroxy group, followed by azidation of the resultant monotosylate afforded a hydroxy azide, which



Scheme 2. Reagents and conditions: a) TsCl, Py, 60%; b) NaN_3, DMF, 80°C, 75%; c) TBSCl, imidazole, 100%; d) aq. NaOH, MeOH, then HOSu, EDCI, 100%; e) **8b**, EtOAc, reflux, 96%; f) Dess–Martin oxidation then $NaClO_2$, resorcinol, NaH_2PO_4, 74%; g) HOSu, EDCI, 85%. EDCI = 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride, Su = succinimide.

was protected with TBSCl to give **10**. After **10** was hydrolyzed with NaOH, the resultant acid was converted into the corresponding activated ester,[11] which when coupled with the amine **8b**, to provide amide **11a** in 96% yield. The hydroxy group in **11a** was oxidated stepwise to the corresponding acid by reaction with the Dess–Martin reagent[12] and sodium chlorite, followed by esterification with HOSu and EDCI[13] to give dipeptide **11b** in 63% yield over three steps; this intermediate was ready for connection with the tetrapeptide part.

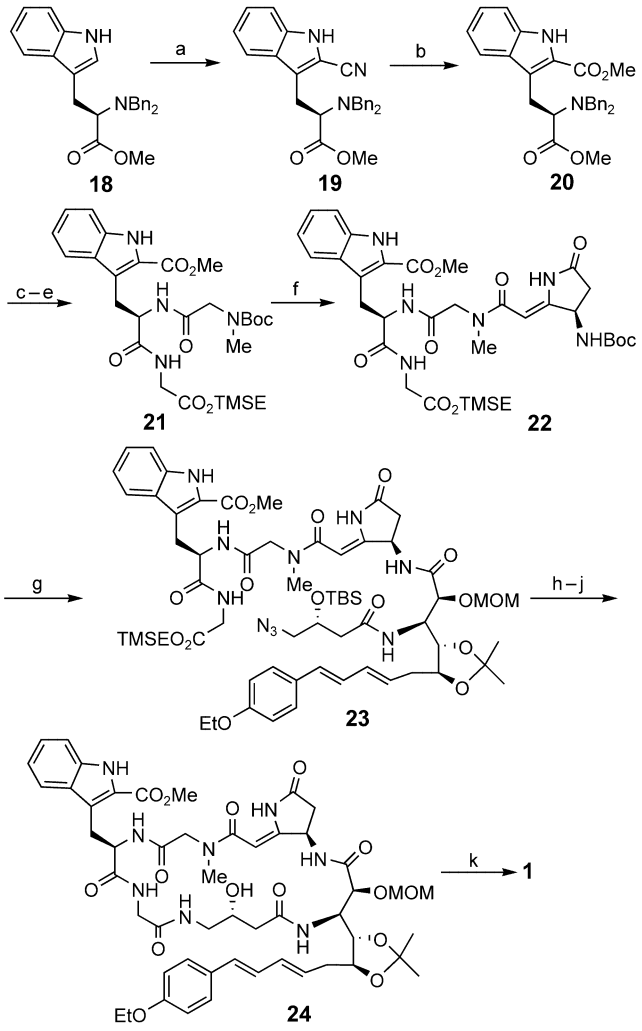
For the synthesis of the pyrrolidinone part, the starting D-aspartic acid derivative **12** was converted into the urethane *N*-carboxyanhydride **14** (Scheme 3),[14] and then treated with the lithium enolate of trimethylsilylethyl (TMSE) acetate to give β-keto ester **15**. This operation is essential for obtaining enantiopure **15** (>98% *ee* determined by HPLC) because attempts to use other activated esters prepared with *N,N*′-carbonyldiimidazole,[4c] *N*-hydroxysuccinimide, and pentafluorophenol led to partial or full racemization of **15**. After hydrogenolysis of the benzyl group over Pd/C, the resultant acid was converted into its mixed anhydride, which was



Scheme 3. Reagents and conditions: a) Allyl bromide, $KHCO_3$, DMF, 97%; b) $(Boc)_2O$, DMAP, 100%; c) $[RhCl(PPh_3)_3]$, 90% EtOH, 81%; d) $SOCl_2$, Py, DMF, 90%; e) $LiCH_2CO_2TMSE$, THF, −78°C, 80%; f) Pd/C, H_2, 100%; g) *i*BuOCOCl, NMM; h) aq NH_3; i) MsCl, Et_3N, 81% for 3 steps; j) TBAF, TsOH then C_6F_5OH, EDCI, 73%. Boc = *tert*-butoxycarbonyl, NMM = *N*-methylmorpholine, TMSE = trimethylsilylethyl.

treated with aqueous ammonia to give a hydroxypyrrolidinone. Subjecting this intermediate to elimination through mesylation provided pyrrolidinone **16** in 81% overall yield. When the TMSE group was removed by treatment with TBAF, the liberated acid was found to be racemized due to the basic conditions directed by TBAF. The racemization could be hampered in the presence of TsOH,[15] although the best *R*/*S* ratio was 4:1. The free acid was converted to its PFP-activated ester[16] **17** by treatment with C_6F_5OH and EDCI in 80% yield.

Construction of the (*R*)-Trp-2-carboxylic acid part commenced with introduction of a cyano group to the indole ring of **18**, which was derived from D-tryptophan according to Danishefsky's protocol[17] (Scheme 4). The cyano group of **19** was hydrolyzed to the acid function under alkali conditions and then esterified to afford the (*R*)-Trp-2-carboxylic acid derivative **20**.



Scheme 4. Reagents and conditions: a) *t*BuOCl, Et_3N, −78 °C then TMSCN, $BF_3{\cdot}Et_2O$, 79%; b) aq KOH, MeOH then MeI, $KHCO_3$, DMF, 71%; c) $Pd(OH)_2$, H_2, MeOH, 70%; d) BocN(Me)CH_2CO_2H, EDCI, HOBt, DIEA, 96%; e) aq LiOH, MeOH then $NH_2CH_2CO_2TMSE$, EDCI, HOBt, DIEA, 88%; f) TFA then **17**, $NaHCO_3$, DMF, 45 °C; 81%; g) TFA then **11b**, $NaHCO_3$, MeCN, 80%; h) TBAF, THF; i) Me_3P, THF; j) DPPA, DMF, 40% for 3 steps; k) LiOH, MeOH, THF, then Amberlyst-15, MeOH, H_2O, 61%. DIEA = diisopropylethylamine, DPPA = diphenylphosphoryl azide, HOBt = 1-hydroxybenzotriazole, TFA = trifluoroacetic acid.

With the four unnatural amino acid fragments in hand, we set out on the next step—coupling these units to complete the synthesis of the macrocyclic core. The reaction was initially carried out on compound **20**. After catalytic removal of the benzyl groups over $Pd(OH)_2$, the free amine was coupled with Boc-*N*-MeGly-OH promoted by EDCI–HOBt[18] to provide a dipeptide (93%). Regioselective hydrolysis of the aliphatic carboxylic ester with LiOH, followed by coupling with H-Gly-OTMSE produced tripeptide **21**. The Boc group was removed by treatment with TFA, and without purification the free amine was coupled with the PFP-activated ester **17** to afford tetrapeptide **22** as a inseparable mixture (4:1), which resulted from the partial racemization of **17**. Removal of the Boc group in **22** followed by coupling with the dipeptide **11b** gave the linear precursor **23** in 80% yield. The TMSE group of **23** was removed with TBAF and TsOH to give the free carboxylic acid. Although attempts to reduce the azide group to the amine failed when **23** was treated with Ph_3P, nBu_3P, and $SnCl_2$,[19] less sterically hindered Me_3P[20] proved to be suitable for this transformation and furnished the reduced product in good yield.

The stage was now set for the crucial macrocyclization. To our delight, although EDCI–HOAt was not effective for this conversion, treatment of the above amino acid with DPPA[21] for 14 days afforded lactam **24** in 40% yield (three steps from **23**), together with its separable isomer (9% yield, resulting from the partial racemization during assembly of the pyrrolidinone fragment). Noteworthy is that much lower yields were observed when the reaction time was shortened to two to three days with either DPPA or HATU[22] as the coupling reagents. Removal of methyl ester was achieved by treatment with LiOH to give the free acid, and the next task was the removal of the MOM and acetonide group. This operation proved far from trivial, mainly because both the substrate and the product are unstable to strong acidic conditions. After some experimentation, we finally observed that the removal was accomplished with Amberlyst-15[23] to give **1**[24] in 61% yield (two steps). The analytical data of the synthetic compound were identical with those reported,[2b] thereby confirming the structure of microsclerodermin E proposed by Faulkner and co-workers.

In summary, we have developed a new, efficient strategy for the enantiospecific synthesis of microsclerodermins exemplified by the first total synthesis of microsclerodermin E (1% overall yield for 26 linear steps). The methods expressed here for the synthesizing AETD and the pyrrolidinone parts should be applicable for installation of the other polyhydroxy-β-amino acid residues and pyrrolidinone units in the microsclerodermins. We are currently extending these methods to the total synthesis of microsclerodermins A–D and their analogues for in-depth biological evaluation. Results will be reported in due course.

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[1] C. A. Bewley, D. J. Faulkner, *Angew. Chem.* **1998**, *110*, 2280; *Angew. Chem. Int. Ed.* **1998**, *37*, 2162.

[2] a) C. A. Bewley, C. Debitus, D. J. Faulkner, *J. Am. Chem. Soc.* **1994**, *116*, 7631; b) E. W. Schmidt, D. J. Faulkner, *Tetrahedron* **1998**, *54*, 3043; c) A. Qureshi, P. L. Colin, D. J. Faulkner, *Tetrahedron* **2000**, *56*, 3679.

[3] A. E. Wright, S. A. Pomponi, R. E. Longley, R. A. Isbrucker, US 6384187, **2002** [*Chem. Abstr.* **2002**, *136*, 319381]

[4] a) S. Sasaki, Y. Hamada, T. Shioiri, *Tetrahedron Lett.* **1997**, *38*, 3013; b) S. Sasaki, Y. Hamada, T. Shioiri, *Tetrahedron Lett.* **1999**, *40*, 3187; c) S. Sasaki, Y. Hamada, T. Shioiri, *Synlett* **1999**, *4*, 453.

[5] S. Marczak, J. Wicha, *Synth. Commun.* **1990**, *20*, 1511.

[6] S. L. Schreiber, H. V. Meyers, *J. Am. Chem. Soc.* **1988**, *110*, 5198.

[7] R. Van Rijsbergen, R. M. J. O. Anteunis, A. De Bruyn, *J. Carbohydr. Chem.* **1983**, *2*, 395.

[8] Z. H. Kudzin, P. Lyzwa, J. Luczak, G. Andrijewski, *Synthesis* **1997**, 44.

[9] S. Saito, T. Hasegawa, M. Inaba, R. Nishida, T. Fujii, S. Nomiz, T. Moriwake, *Chem. Lett.* **1984**, 1389.

[10] F. D. Bellamy, M. Bondoux, P. Dodey, *Tetrahedron Lett.* **1990**, *31*, 7323.

[11] E. Wunsch, E. Drees, *Chem. Ber.* **1966**, *99*, 110.

[12] D. B. Dess, J. C. Martin, *J. Org. Chem.* **1983**, *48*, 4155.

[13] J. C. Sheehan, P. A. Cruickshank, G. L. Boshart, *J. Org. Chem.* **1961**, *26*, 2525.

[14] M. Paris, J. Fehrentz, A. Heitz, A. Loffet, J. Martinez, *Tetrahedron Lett.* **1996**, *37*, 8489.

[15] J. P. Ferezou, M. Julia, Y. Li, L. W. Liu, A. Pancrazi, *Bull. Soc. Chim. Fr.* **1995**, *132*, 428.

[16] U. Schmidt, M. Kroner, H. Griesser, *Synthesis* **1989**, 832.

[17] J. M. Schkeryantz, J. G. Woo, P. Siliphaivanh, K. M. Depew, S. J. Danishefsky, *J. Am. Chem. Soc.* **1999**, *121*, 11969.

[18] W. König, R. Geiger, *Chem. Ber.* **1970**, *103*, 788.

[19] E. F. V. Scriven, K. Turnbull, *Chem. Rev.* **1988**, *88*, 297.

[20] B. M. Trost, S. R. Pulley, *Tetrahedron Lett.* **1995**, *36*, 8737.

[21] a) D. L. Boger, P. Yohannes, *Tetrahedron Lett.* **1989**, *30*, 5061; b) T. Sugiura, Y. Hamada, T. Shioiri, *Tetrahedron Lett.* **1987**, *28*, 2251; c) M. P. Forst, G. P. Concepcion, G. B. Caraan, C. M. Ireland, *J. Org. Chem.* **1992**, *57*, 6671.

[22] L. A. Carpino, A. El-Faham, C. A. Minor, F. Albericio, *J. Chem. Soc. Chem. Commun.* **1994**, 201.

[23] B. M. Trost, J. D. Chisholm, S. T. Wrobleski, M. Jung, *J. Am. Chem. Soc.* **2002**, *124*, 12420.

[24] $[\alpha]_D^{25} = -22.7$ ($c = 0.4$, 1:1 MeOH/0.1N NH_4HCO_3 (aq); lit.[2b]: $[\alpha]_D^{25} = -24$ ($c = 0.4$, 1:1 MeOH/0.1N NH_4HCO_3 (aq)); ^{1}H NMR (300 MHz, [D$_7$]DMF with trace of TFA): $\delta = 11.58$ (s, 1H), 10.60 (s, 1H), 8.88 (d, $J = 4.8$ Hz, 1H), 8.62 (d, $J = 8.7$ Hz, 1H), 8.49 (t, $J = 6.2$ Hz, 1H), 7.72 (d, $J = 7.8$ Hz, 1H), 7.59 (br, 1H), 7.54 (d, $J = 8.1$ Hz, 1H), 7.40 (d, $J = 8.7$ Hz, 2H), 7.35 (m, 1H), 7.31 (t, $J = 7.8$ Hz, 1H), 7.16 (t, $J = 7.8$ Hz, 1H), 6.91 (d, $J = 8.7$ Hz, 2H), 6.73 (dd, $J = 15.9$, 10.8 Hz, 1H), 6.45 (d, $J = 15.9$ Hz, 1H), 6.24 (dd, $J = 15.0$, 10.8 Hz, 1H), 5.83 (m, 1H), 5.45 (s, 1H), 5.42 (m, 1H), 4.68 (s, 1H), 4.64 (d, $J = 15.9$ Hz, 1H), 4.33 (m, 1H), 4.27 (m, 1H), 4.05 (q, $J = 5.7$ Hz, 2H), 3.95 (br, 1H), 3.73 (m, 1H), 3.68 (m, 2H), 3.61 (m, 2H), 3.47 (d, $J = 15.9$ Hz, 1H), 3.45 (m, 2H), 3.07 (s, 3H), 2.91 (m, 2H), 2.59 (dd, $J = 18.9$, 4.8 Hz, 1H), 2.48 (d, $J = 13.5$ Hz, 1H), 2.39 (m, 2H), 2.21 (m, 1H), 1.35 ppm (t, $J = 5.7$ Hz, 3H); negative ESI-MS m/z 929 $(M-H)^-$.

