Total Synthesis of Cryptophycins and Their 16-(3-Phenylacryloyl) Derivatives

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Cryptophycin A, a cyclic depsipeptide isolated from the blue-green alga (cyanobacterium) *Nostoc* sp.GSV 224, has shown excellent activity against solid tumors implanted in mice. The benzylic epoxide, which was shown to be very important for biological activity, is also fairly unstable under both acidic and alkaline conditions. The high doses needed to observe *in vivo* activity might be a result of this instability. In order to solve this problem while preserving the electrophilic character of the benzylic position, enones **1** and **2** have been proposed as promising analogs of the natural product, and a convergent total synthesis of these compounds is described. In addition, the same strategy was used to prepare Cryptophycins A, B, C, and D.

In 1994, Moore and co-workers isolated from the blue-green alga (cyanobacterium) *Nostoc* sp. GSV 224, a series of metabolites known as Cryptophycins (Figure 1).[1] These compounds displayed very potent *in vitro* cytotoxicity against several human tumor cell lines and experimental evidence shows that the effect is due to irreversible inhibition of tubulin polymerization into microtubules.[2] Both *in vitro*[1] and *in vivo*[3] data suggest that the presence of the benzylic epoxide moiety is very important for biological activity. For instance, Cryptophycin A shows a much better *in vitro* profile than its deoxy counterpart Cryptophycin C in the KB (human nasopharyngeal cell line)(IC_{50} A: 5 pg/mL; IC_{50} C: 3000 pg/mL).[1] The *in vivo* studies follow the same trend but despite its very high *in vitro* potency, Cryptophycin A displays very good activity only at relatively high doses (30−132 mg/kg).[3] One of the possible reasons for this poor correlation may be the inherent instability of the benzylic epoxide which might generate inactive compounds. In order to solve this problem while preserving the electrophilic character of the benzylic position, enones **1** and **2** were envisioned as promising analogs of the natural products.

Retrosynthetic analysis of **2** led to four subunits (A−D).[4] D-tyrosine (B) and (*S*)-(−)-2-hydroxyisocaproic acid (D) are commercially available. Since the *β*-amino acid subunit (C) is available only in its racemic form, a synthetic route to the desired enantiomer was developed. Finally, an enantioselective synthesis of fragment A was also elaborated. Subunits B, C, and D were preassembled, and coupling with A was done through ester bond formation between A and D. Cyclization was then carried out by a macrolactamization reaction between A and B.

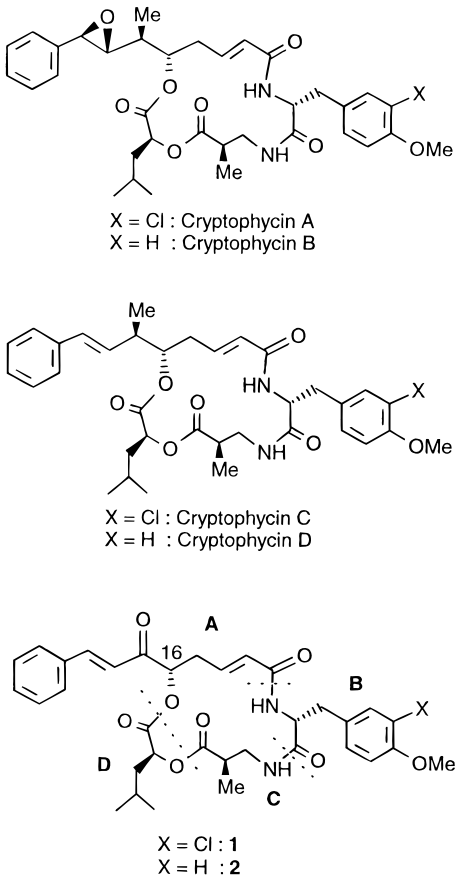


Figure 1.

The synthesis of fragment A (Scheme 1) was realized in five practical steps. Phenylacetylene (**3**) was treated with *n*-butyllithium and added to a solution of (*S*)-(−)-2-acetoxysuccinic anhydride[5] in tetrahydrofuran at −78 °C. After 10 min, sodium borohydride was added, followed by an aqueous solution of sodium hydroxide, to produce a diastereomeric mixture of carboxylic acids **4**. Interestingly, the anhydride opening was regioselective,[6] whereas the ketone reduction led a 1:1 mixture of both isomers. Heating this mixture with *p*-toluenesulfonic

[⊗] Abstract published in *Advance ACS Abstracts,* August 1, 1996.

(1) Trimurtulu, G.; Ohtani, I.; Patterson, G. M. L.; Moore, R. E.; Corbett, T. H.;Valeriote, F. A.; Demchik, L. *J. Am. Chem. Soc.* **1994**, *116*, 4729. Cryptophycin A was first isolated from *Nostoc* sp. ATCC 53789 by researchers at Merck. See: Schwartz, R. E.; Hirsch, C. F.; Sesin, D. F.; Flor, J. E.; Chartrain, M.; Fromtling, R. E.; Harris, G. H.; Salvatore, M. J.; Liesch, J. M.; Yudin, K. *J. Ind. Microbiol.* **1990**, *5*, 113.

(2) Smith, C. D.; Zhang, X.; Mooberry, S. L.; Patterson, G. M. L.; Moore, R. E. *Cancer Res.* **1994**, *54*, 3779.

(3) Trimurtulu, G.; Ogino, J.; Heltzel, C. E.; Husebo, T. L.; Jensen, C. M.; Larsen, L. K.; Patterson, G. M. L.; Moore, R. E.; Mooberry, S. L.; Corbett, T. H.;Valeriote, F. A. *J. Am. Chem. Soc.* **1995**, *117*, 12030.

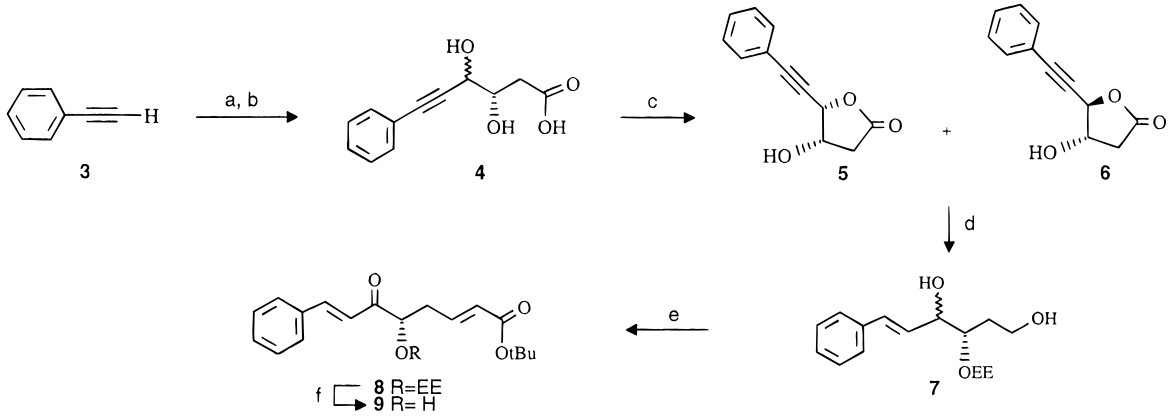
(4) Barrrow, R. A.; Hemscheidt, T.; Liang, J.; Paik, S.; Moore, R. E.; Tius, M. A. *J. Am. Chem. Soc.* **1995**, *117*, 2479.

(5) Commercially available from Aldrich Co.

(6) The same regioselectivity was observed with an alcohol as nucleophile; See: Shiuey, S.-J.; Partridge, J. J.; Uskokovic, M. R. *J. Org. Chem.* **1988**, *53*, 1040.

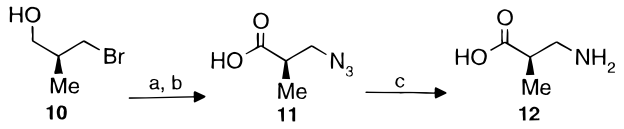
S0022-3263(96)00816-X CCC: $12.00

Scheme 1[a]



[a] (a) BuLi, THF, −78 °C; then (S)-(−)-2-acetoxysuccinic anhydride; (b) $NaBH_4$, EtOH, −78 °C; then NaOH 0 °C; PTSA, benzene, 50 °C, 63% from **3**; (d) ethyl vinyl ether, PTSA, ether, 0 °C to rt; then LAH, 0 °C to rt; (e) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, −78 °C to 0 °C; then (*tert*-butoxycarbonylmethylene)triphenylphosphorane, 65% from **6**; (f) AcOH, H_2O, rt, 73%.

Scheme 2[a]



[a] (a) NaN_3, DMF, 100 °C; (b) Jones reagent, acetone, rt, 93% from **10**; (c) Pd/C, H_2, EtOAc, EtOH, rt, 70%.

acid (PTSA) in benzene at 50 °C, afforded butyrolactones **5** and **6**, easily separated by flash chromatography, in 63% combined yield from **3**. Both lactones could be used to complete the synthesis of fragment A. For example, **6** was treated with ethyl vinyl ether and a catalytic amount of *p*-toluenesulfonic acid in ether. After 1 h, lithium aluminum hydride was added, and the reaction mixture was then stirred at room temperature for 24 h. The resulting diol **7** was immediately treated with 2.5 equiv of Swern reagent followed by 1.4 equiv of (*tert*-butoxycarbonylmethylene)triphenylphosphorane to give **8** in 65% overall from **6**. The ethoxy ethyl group was then cleaved in acetic acid and water affording alcohol **9** in 73% yield after trituration of the crude product in hexanes.

The *β*-amino acid **12** (fragment C, Scheme 2), was prepared from the commercially available (*S*)-(+)-3-bromo-2-methyl-1-propanol[7] (**10**) in three steps. The bromide was displaced with sodium azide in dimethylformamide at 100 °C, and the resulting azido alcohol was oxidized with Jones reagent in acetone to produce the carboxylic acid **11** in 93% overall yield from **10**. Subsequent hydrogenation over palladium afforded the *β*-amino acid **12** in 70% yield.

Having the four subunits in hand, the synthesis of **2** (Scheme 3) was undertaken. The use of *N*-hydroxysuccinimide esters[8] as activated intermediates, allowed us to prepare the (BCD)-fragment in two steps, minimizing the use of protecting groups. Thus, N-protected D-tyrosine (**13**) was treated with dicyclohexylcarbodiimide (DCC) and *N*-hydroxysuccinimide in dry dimethoxyethane at 0 °C. Then **12** and triethylamine in water were added to the reaction mixture affording, after workup, adduct **14** in 93% yield as a single diastereomer (>95% de by ^1H NMR). This product was reacted with 1-[3-(dimethylamino)propyl]-3-ethylcarbodiimide hydrochloride (EDCI) and *N*-hydroxysuccinimide in dimethoxyethane and dichloromethane. The activated ester was then treated with (*S*)-(−)-2-hydroxyisocaproic acid and (dimethylamino)pyridine (DMAP) in acetonitrile to produce the fragment (BCD) **15** in 85% yield after purification. The coupling reaction between fragments A and (BCD) was carried out with trichlorobenzoyl chloride, triethylamine, and DMAP[9] to produce **16** in 84% yield and >95% de. The protecting groups were then cleaved by a treatment with trifluoroacetic acid in dichloromethane. The resulting amino acid was cyclized with *O*-benzotriazol-1-yl-*N*,*N*,*N*′,*N*′-bis(pentamethylene)uronium hexafluorophosphate[10] and diisopropylethylamine in acetonitrile at 0 °C. Compound **2** was obtained in 69% yield from **16**. Due to some epimerization during the deprotection or the cyclization step, 5−10% of the 16-epi diastereomer was isolated from the reaction mixture. The macrocyclic structure **2** was prepared in five steps and 46% overall yield from the four subunits A−D.

In order to validate our synthetic route and generate positive controls for the biological evaluation of our analogs, we used our synthetic strategy to prepare Cryptophycins A−D. The A fragment of these compounds could be obtained independently from either butyrolactone **5** or its epimer **6** via separate routes (Schemes 4 and 5). In the first approach, butyrolactone **5** (Scheme 4), was treated with dihydropyran and PTSA, in tetrahydrofuran, followed by lithium aluminum hydride (LAH) reduction in ether affording diol **17** in 92% overall yield. The primary alcohol was selectively protected as a pivaloyl ester, and then the secondary hydroxyl was acetylated (59% overall): The dimethyl cuprate reaction on the fully protected compound failed and led to a mixture of conjugated dienes (results not shown), probably by a reduction−elimination mechanism. In order to avoid this process, the tetrahydropyranyl group was cleaved and the reaction was carried out on **18**. In this case, the reaction led to a 1:1 mixture of S_N2 and S_N2' products from which the desired product **19** was isolated by flash chromatography in 34% yield. The secondary alcohol was reprotected as tetrahydropyranyl

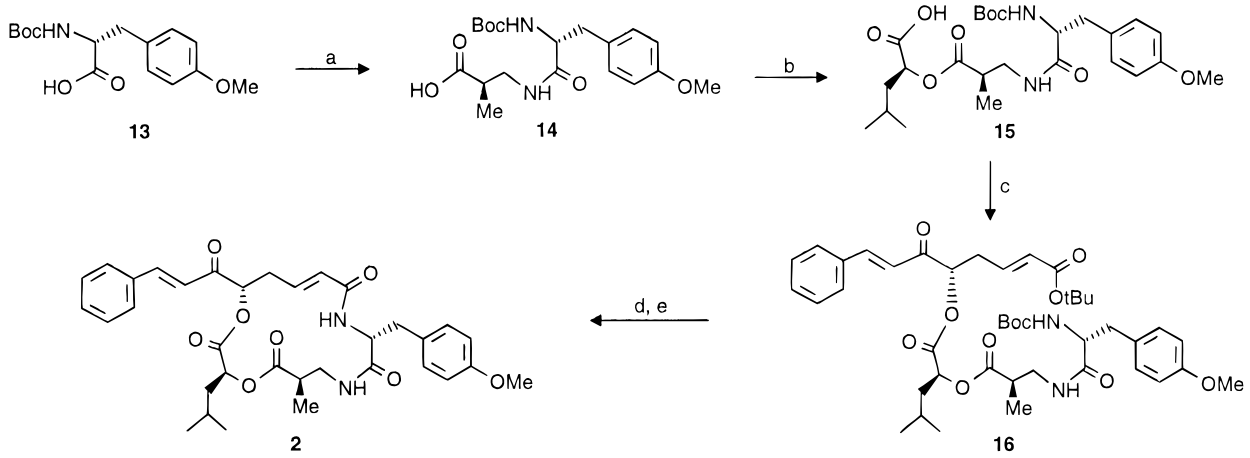
(7) **10** could also be prepared by reduction of methyl (*S*)-(+)-3-bromo-2-methyl propionate with LAH in ether at −50 °C.

(8) Anderson, G. W.; Zimmerman, J. E.; Callahan, F. M. *J. Am. Chem. Soc.* **1963**, *85*, 3039.

(9) Inanaga, J.; Hirata, K.; Saeki, H. Katsuki, T.; Yamaguchi, M. *Bull. Chem. Soc. Jpn.* **1979**, *52*, 1989.

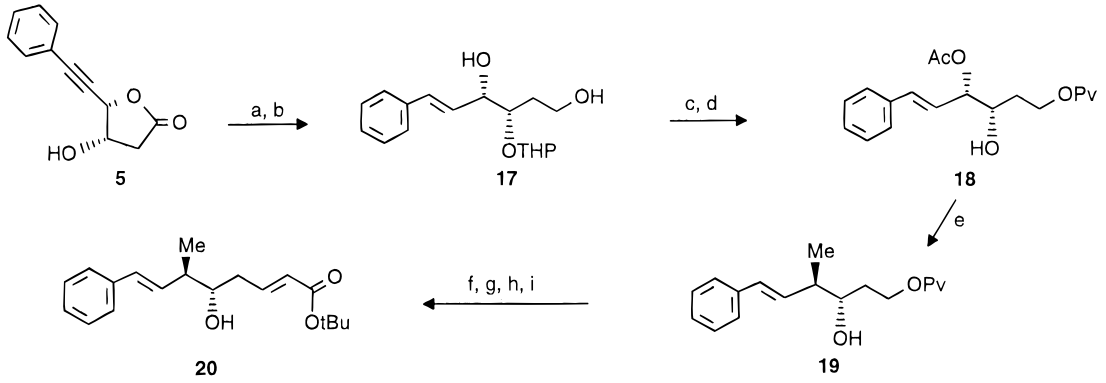
(10) Ehrlich, A.; Rothemund, S.; Brudel, M.; Beyermann, M.; Carpino L. A.; Bienert, M. *Tetrahedron Lett.* **1993**, *30*, 4781.

Scheme 3[a]



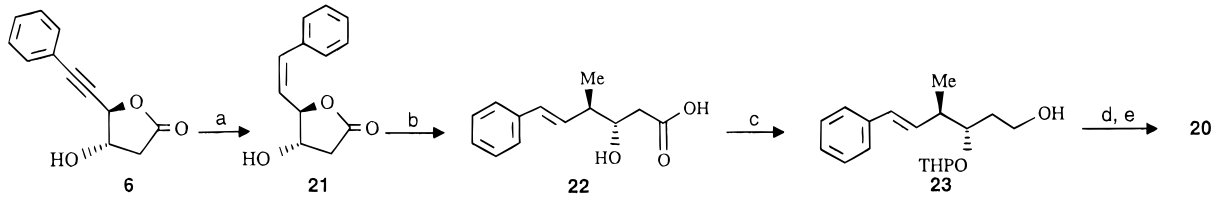
[a] (a) DCC, N-hydroxysuccinimide, DME, 0 °C; then 12, Et_3N, H_2O, rt, 93%; (b) EDCl, *N*-hydroxysuccinimide, DME, CH_2Cl_2; then (S)-(−)-2-hydroxyisocaproic acid; DMAP, CH_3CN, rt, 85%; (c) trichlorobenzoyl chloride, Et_3N, THF, rt, 84%; (d) TFA, CH_2Cl_2; (e) *O*-benzotriazol-1-yl-*N*,*N*,*N*′,*N*′-bis(pentamethylene)uronium hexafluorophosphate, DIPEA, CH_3CN, 0 °C, 69% from **16**.

Scheme 4[a]



[a] (a) DHP, PTSA, THF, rt, 92%; (b) LAH, Et_2O, rt, 100%; (c) PvCl, pyr, DMAP, 0 °C to rt; then Ac_2O, 59%; (d) AcOH, H_2O, 100%; (e) $(Me)_2CuLi$, Et_2O, 0 °C, 34%; (f) DHP, PTSA, THF, rt, 98%; (g) LAH, THF, 0 °C, 97%; (h) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, −78 °C to 0 °C; then (*tert*-butoxycarbonylmethylene)triphenylphosphorane, 73% (for two steps); (i) AcOH, H_2O, 40 °C, 80%.

Scheme 5[a]



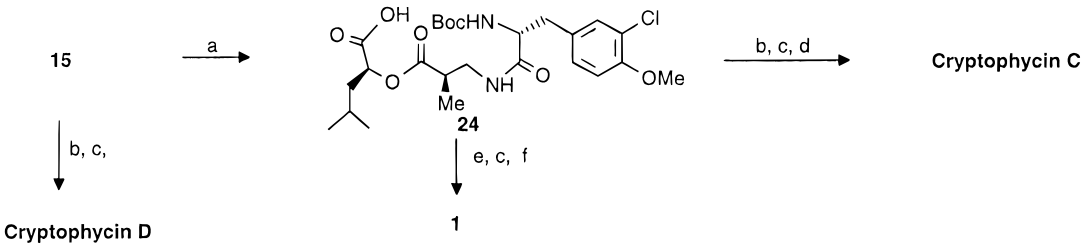
[a] (a) H_2, Lindlar catalyst, quinoline, EtOAc, MeOH, 0 °C, 85%; (b) CuBr $(Me)_2S$, MeLi, Et_2O, −35 °C; (c) DHP, PTSA, THF, rt, then LAH, 0 °C; (d) $(COCl)_2$, DMSO, Et_3N, CH_2Cl_2, −78 °C to 0°; then (*tert*-butoxycarbonylmethylene)triphenylphosphorane; (e) AcOH, H_2O, 40 °C, 23% from 21.

derivative, and the pivaloate group was then cleaved with LAH. Swern oxidation of the resulting alcohol followed by reaction with (*tert*-butoxycarbonylmethylene)triphenylphosphorane afforded, after treatment with acetic acid and water, the desired alcohol **20** in 58% overall yield from **19**. The second approach starts with butyrolactone **6** (Scheme 5), which was partially hydrogenated over Lindlar catalyst, to produce **21** in 85% yield. Addition of dimethyl cuprate on **21** resulted in retention of stereochemistry at the electrophilic center and complete isomerization of the double bond.[11] The S_N2' product, also present in this case, was separated only at the last step. The sequence was completed in a similar manner as described for the previous approach. Protection of the hydroxyl group followed by reduction of the carboxylic acid afforded alcohol **23**. Swern oxidation followed by Wittig reaction and finally deprotection of the secondary alcohol afforded **20** in 23% overall yield from **21**.

Chlorination of the aromatic ring of the tyrosine moiety was needed in order to prepare Cryptophycins A and C as well as our enone analog **1**. Introduction of the chlorine atom could be done directly on the BCD fragment **15** (Scheme 6) by treament with sulfuryl chloride in acetic acid,[4] followed by BOC-ON and triethylamine to reprotect the amino group. The chloro derivative **24** was obtained in 68% yield from **15**. The synthesis of **1** and Cryptophycin C was easily completed by first cou-

(11) Goering, H. L.; Kantner, S. S. *J. Org. Chem.* **1984**, *49*, 422.

Scheme 6[a]



[a] (a) SO_2Cl_2, AcOH, 55 °C; then BOC-ON, Et_3N, CH_2Cl_2 68%; (b) trichlorobenzoyl chloride, Et_3N, THF, rt; then 20, DMAP, Et_3N, toluene, rt, 58−68%; (c) TFA, CH_2Cl_2; (d) *O*-benzotriazol-1-yl-*N*,*N*,*N*′,*N*′-bis(pentamethylene)uronium hexafluorophosphate, DIPEA, DMF, 0 °C, 85% for two steps; (e) condition b with **9** instead of **20**; (f) condition d, 51% from **24**; (g) condition d with DMF replaced by acetonitrile, 35% from 15.

pling **24** with the appropriate subunit A (**9** or **20**). The protecting groups were then cleaved, and the resulting acyclic precursors were cyclized using the same procedure as described for enone **2**. Cryptophycin D was obtained by coupling **15** with **20**, followed by the usual deprotection and cyclization steps. Finally, Cryptophycins A and B were prepared from Cryptophycins C and D, respectively, by epoxidation with *m*-chloroperbenzoic acid[4] or dimethyldioxirane.[12] The spectroscopic data and the biological activities of all synthetic Cryptophycins are in perfect agreement with the reported values.[1,4]

Conclusion

In summary, we have developed a convergent total synthesis of Cryptophycins and their 16-(3-phenylacryloyl) derivatives. The use of *N*-hydroxysuccinimide esters as activated intermediates allowed us to minimize the use of protecting groups. Compounds **1** and **2**, where the benzylic epoxide of Cryptophycins A and B was replaced by an enone moiety, were found to be very poor cytotoxic agents.[13] Consequently, the biological activity of Cryptophycin A is not solely related to the electrophilic character of the benzylic position.

Experimental Section

General Methods. Unless otherwise noted, all common reagents and solvents were used as obtained from commercial suppliers without further purification. Elemental analysis were carried out by Canadian Microanalytical Service Ltd., Delta, BC. Melting points were determined on a Digital Melting Point Apparatus and are uncorrected. Merck precoated silica gel 60 F254 plates were used for thin-layer chromatography (TLC). EM Science silica gel 60 (230−400 mesh ASTM) was used for flash chromatography. All reactions requiring anhydrous conditions were conducted under a positive pressure of nitrogen.

(3*S*,4*S*) and (3*S*,4*R*)-3,4-Dihydroxy-6-phenylhex-5-ynoic Acid (4). To a stirred solution of (*S*)-(−)-2-Acetoxysuccinic anhydride (10 g, 63.2 mmol) in THF (50 mL) at −78 °C was added dropwise during 25 min a freshly prepared solution of lithiated phenylacetylene (prepared by treating phenylacetylene (6.38 mL, 57.0 mmol) with *n*-butyllithium (2.5 M, 23 mL) at −78°C in THF (40 mL)). After the addition, the mixture was stirred for 10 min. A solution of sodium borohydride (2.5 g) in ethanol (50 mL) was slowly added to the reaction mixture at −78°C. After 15 min, water (5 mL) was added followed by a solution of 1 N NaOH (63 mL). The reaction mixture was warmed up to 0°C, and after 20 min, carefully acidified to pH 4 with 3 N HCl (and 1 N HCl toward the end). The mixture was extracted with ethyl acetate (3 × 300 mL), and the combined organic phases were washed with water and brine and then dried over Na_2SO_4 and evaporated to give 10.1 g (73%) of **4**. ^{1}H NMR (400 MHz, $CDCl_3$) 7.45 (m, 2H), 7.34 (m, 3H), 4.73 and 4.56 (2d, J = 3.9 and 6.3 Hz, 1H), 4.28 and 4.23 (2m, 1H), 2.88 and 2.73 (2m, 2H); HRMS (M + NH_4^+) calcd for $C_{12}H_{16}NO_4$: 238.1079, found: 238.1078.

(4*S*,5*S*) and (4*S*,5*R*) 4-Hydroxy-5-(phenylethynyl)dihydrofuran-2-one (5 and 6). A stirred mixture of **4** (5.0 g, 22.7 mmol) and PTSA (200 mg, 1.0 mmol) in benzene (300 mL) was heated to 50 °C for 1.5 h. The mixture was cooled and neutralized with pyridine (85 mL), and then the solvent was evaporated. The residue was purified by flash chromatography (hexane:EtOAc 3:2) to give **5** (2.0 g, 43%) and **6** (2.1 g, 45%). Compound **5**: mp: 133−135 °C; $[\alpha]_D$ −52.2° (*c* 0.5, $CHCl_3$); IR (neat film) 3400, 1795, 1156 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 7.51 (m, 2H), 7.40 (m, 3H), 5.36 (d, J = 3.9 Hz, 1H), 4.63 (m, 1H), 2.80 (dd, J = 5.2, 17.6 Hz, 1H), 2.73 (dd, J = 2.4, 17.6 Hz, 1H), 2.62 (d, J = 2.5 Hz, 1H); ^{13}C NMR (75 MHz, $CDCl_3$) 174.1, 132.0, 129.6, 128.5, 120.7, 91.4, 79.7, 75.3, 68.7, 37.1; HRMS (M^+) calcd for $C_{12}H_{10}O_3$: 202.0630, found: 202.0637. Anal. Calcd for $C_{12}H_{10}O_3$: C, 71.28; H, 4.98. Found: C, 71.16; H, 5.05. Compound **6**: mp 66−68 °C; $[\alpha]_D$ +53.6° (*c* 0.5, $CHCl_3$); IR (neat film) 3430, 1780, 1154 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 7.45 (m, 2H), 7.37 (m, 3H), 5.23 (d, J = 1.4 Hz, 1H), 4.74 (m, 1H), 3.04 (dd, J = 5.9, 17.7 Hz, 1H), 2.58 (dd, J = 2.4, 17.6 Hz, 1H), 2.24 (d, J = 4.2 Hz, 1H); ^{13}C NMR (75 MHz, $CDCl_3$) 174.7, 131.8, 129.3, 128.4, 121.1, 89.1, 82.2, 76.7, 73.4, 36.9; HRMS (M^+) calcd for $C_{12}H_{10}O_3$: 202.0630, found: 202.0637. Anal. Calcd for $C_{12}H_{10}O_3$: C, 71.28; H, 4.98. Found: C, 71.44; H, 5.08.

(5*S*)-5-Hydroxy-6-oxo-8-phenylocta-2(*E*),7(*E*)-dienoic Acid *tert*-Butyl Ester (9). To a stirred solution of butyrolactone **6** (519 mg, 2.57 mmol) in ether (25 mL) were added ethyl vinyl ether (918 mL, 9.60 mmol) and PTSA (17 mg, 0.09 mmol) at 0 °C. After 30 min, the reaction mixture was warmed up to room temperature and stirred for 1 h. Then lithium aluminum hydride (273 mg, 7.2 mmol) was added at 0 °C, and the reaction mixture was stirred at room temperature for 24 h. Methanol (1 mL) was then added, and the reaction mixture was worked up in ethyl acetate and 5% $NaHCO_3$. The solvents were evaporated, and the crude diol **7** was dissolved in CH_2Cl_2 (4.5 mL) and added to a solution of DMSO (933 mL, 13.1 mmol) and $(COCl)_2$ (573 mL, 6.57 mmol) in CH_2Cl_2 (20 mL) at −78 °C. After 20 min, triethylamine (2.7 mL, 19.4 mmol) was added and after a few min, the temperature was raised to 0 °C and (*tert*-butoxycarbonylmethylene)triphenylphosphorane (1.40 g, 3.72 mmol) was added. After 15 min, the solvent was evaporated, and the product was purified by flash chromatography (hexane−EtOAc 8:1) to give **8** (621 mg, 65% from **6**). The ethoxy ethyl derivative **8** (608 mg, 1.62 mmol) was dissolved in acetic acid (4 mL) and water (2 mL). After 1 h at room temperature, the reaction mixture was extracted in CH_2Cl_2 and water. The crude product was purified by trituration in hexane to give **9** (356 mg, 73%) as a beige solid: mp 72.5−73.5 °C; $[\alpha]_D$ −80.2° (*c* 1.21, $CHCl_3$); IR (neat film) 3450, 2950, 1711, 1690, 1656, 1610, 1154 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 7.76 (d, J = 16.0 Hz, 1H), 7.58 (m, 2H), 7.43 (m, 3H), 6.85 (d,

(12) Kobayashi, M.; Kurosu, M.; Wang, W.; Kitagawa, I. *Chem. Pharm. Bull.* **1994**, *42*, 2394.

(13) De Muys, J.-M.; Rej, R.; Nguyen, D.; Go, B.; Fortin, S.; Lavallée, J.-F. *BioMed. Chem. Lett.* **1996**, *6*, 1111.

J = 16.0 Hz, 1H), 6.84 (m, 1H), 5.86 (d, J = 15.7 Hz, 1H), 4.58 (dd, J = 4.4, 7.2 Hz, 1H), 3.71 (broad singlet, 1H), 2.79 (m, 1H), 2.51 (m, 1H), 1.49 (s, 9H); ^{13}C NMR (75 MHz, $CDCl_3$) 199.1, 165.2, 145.4, 141.4, 133.8, 131.3, 129.1, 128.7, 126.4, 120.1, 80.4, 74.4, 37.0, 28.1; HRMS (MH^+) calcd for $C_{18}H_{23}O_4$: 303.1596, found: 303.1603. Anal. Calcd for $C_{18}H_{22}O_4$: C, 71.50; H, 7.33. Found: C, 71.26; H, 7.19.

(2*R*)-3-Azido-2-methylpropionic Acid (11). (*S*)-(+)-3-bromo-2-methyl-1-propanol (9.52 g, 62.3 mmol) and NaN_3 (8.1 g, 125 mmol) were heated in DMF (65 mL) at 90−100°C for 3 h. The reaction was worked up by cooling down to room temperature adding ether, filtering off the solid, and washing the filtrate several times with water. The ether phase was washed with brine, dried over $MgSO_4$, and evaporated to give 6.71 g (94%). The residue was then dissolved in acetone (270 mL), and Jones reagent was added in portions until the mixture permanently turned orange. 2-propanol was added to quench excess of reagent. The precipitate was filtered off and rinsed with ether. The filtrate was then washed with brine and dried over $MgSO_4$. Evaporation of the solvent gave 7.50 g (100%) of azido acid **11**: ^{1}H NMR (400 MHz, $CDCl_3$) 3.59 and 3.45 (2dd, J = 12.2 Hz, 7.1 Hz, 2H), 2.75 (m, 1H), 1.28 (d, J = 7.2 Hz, 3H).

(2*R*)-3-Amino-2-methylpropionic Acid (12). A solution of azido acid **11** (7.5 g, 58.1 mmol) and Pd/C 10% (2 g) was stirred under H_2 in EtOAc (200 mL) and EtOH (50 mL) at room temperature for 48 h. The mixture was then filtered over Celite. The first organic filtrate was separated and discarded. The filtration pad was rinsed with water and the aqueous filtrate evaporated to dryness. The crude solid residue was recrystallized from methanol and EtOAc yielding 4.17 g (70%) of the desired β-amino acid **12** as a colorless solid: mp 179−181 °C; $[\alpha]_D$ −14.7° (*c* 2.60, H_2O); ^{1}H NMR (400 MHz, CD_3-OD) 4.9 (bs, 3H), 2.95 (m, 2H), 2.48 (m, 1H), 1.2 (d, J = 7.2 Hz, 3H); ^{13}C NMR (75 MHz, CD_3OD) 179.0, 41.6, 37.7, 13.9; HRMS (M^+) calcd for $C_4H_9NO_2$: 103.0633, found: 103.0631. Anal. Calcd for $C_4H_9NO_2$: C, 46.59; H, 8.80; N, 13.58. Found: C, 46.44; H, 8.73; N, 13.34.

3-[[2(*R*)-(*tert*-Butoxycarbonylamino)-3-(4-methoxyphenyl)propionyl]amino]-2(*R*)-methylpropionic Acid (14). To a stirred solution of Boc-D-Tyr(Me)OH (5.00 g, 16.9 mmol) and *N*-hydroxysuccinimide (2.92 g, 25.4 mmol) in DME (130 mL) at 0 °C was added DCC (4.37 g, 21.2 mmol). After 36 h, a solution of (*R*)-(−)-3-amino-2-methylpropionic acid (2.10 g, 20.3 mmol) and TEA (4.72 mL, 34 mmol) in water (50 mL) was added, and the reaction mixture was warmed up to room temperature. After 30 min, the solid was filtered and rinsed with water. The filtrate was evaporated, and an aqueous solution of 5% K_2CO_3 was added to the residue until pH 9 and extracted with CH_2Cl_2. The aqueous phase was acidified to pH 2 and extracted with CH_2Cl_2. The CH_2Cl_2 phase was then washed with brine, dried over $MgSO_4$, and evaporated to give an oily residue. Ether was added and **14** (6.02 g, 93%) precipitated out as a colorless solid: mp 119−122 °C; $[\alpha]_D$ −6.9° (*c* 1.00, H_2O); IR (neat film) 3314, 2980, 2940, 1710, 1660, 1514, 1465, 1250 cm^{-1}; ^{1}H NMR (400 MHz, acetone-d_6) 10.8 (broad s, 1H), 7.28 (broad s, 1H), 7.16 (d, J = 8.6 Hz, 2H), 6.83 (d, J = 8.6 Hz, 2H), 5.99 (bd, J = 8.6 Hz, 1H), 4.26 (m, 1H, 1H), 3.75 (s, 3H), 3.44 (m, 1H), 3.27 (m, 1H), 3.06 (dd, J = 5.6, 13.8 Hz, 1H), 2.85 (m, 1H), 2.63 (m, 1H), 1.35 (s, 9H), 1.10 (d, J = 7Hz, 3H); ^{13}C NMR (75 MHz, acetone-d_6) 174.7, 170.9, 157.6, 154.4, 129.3, 128.7, 112.7, 77.5, 55.3, 53.6, 40.5, 38.1, 37.1, 26.7, 13.3; HRMS (MH^+) calcd for $C_{19}H_{29}N_2O_6$: 381.2025, found: 381.2023. Anal. Calcd for $C_{19}H_{28}N_2O_6$: C, 59.99; H, 7.42; N, 7.36. Found: C, 59.77; H, 7.19; N, 7.37.

2(*S*)-[[3-[[2(*R*)-(*tert*-Butoxycarbonylamino)-3-(4-methoxyphenyl)propionyl]amino]-2(*R*)-methylpropionyl]oxy]-4-methylpentanoic Acid (15). To a stirred solution of **14** (100 mg, 0.26 mmol) and *N*-hydroxysuccinimide (61 mg, 0.53 mmol) in DME/CH_2Cl_2 (1.5 mL/1.5 mL) was added 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (66 mg, 0.34 mmol) at room temperature. After 24 h, EtOAc was added, and the solution was washed with water, 0.1 N HCl, 5% $NaHCO_3$, water, and brine. The organic phase was then dried over $MgSO_4$ and the solvent evaporated to give the activated hydroxysuccinimide ester (123 mg). The intermediate was next dissolved in CH_3CN (2.6 mL), and DMAP (135 mg, 1.1 mmol) and (2*S*)-2-hydroxy-4-methylpentanoic acid (87 mg, 0.66 mmol) were then added at room temperature. After 15 h, 0.1 N HCl was added to the reaction mixture until pH 2, and then the solution was extracted with EtOAc. The organic phase was washed with water and brine, and dried over $MgSO_4$. After evaporation of the solvents, the residue was purified by flash chromatography (CH_2Cl_2:MeOH:EtOAc 90:5:5) to give **15** (110 mg, 85%) as a ligh beige solid: mp 100−110 °C; $[\alpha]_D$ −21.0° (*c* 4.8, MeOH); IR (neat film) 3315, 2960, 1730, 1668, 1612, 1514, 1463, 1370, 1250 cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6) 8.78 (broad s, 1H), 7.13 (d, J = 8.5 Hz, 2H), 6.90 (d, J = 8.7 Hz, 1H), 6.79 (d, J = 8.5 Hz, 2H), 4.81 (dd, J = 3.7, 9.2 Hz, 1H), 4.04 (m, 1H), 3.70 (s, 3H), 3.24 (bt, J = 5.0 Hz, 2H), 2.82 (dd, J = 4.8 Hz, 13.7 Hz, 1H), 2.66 (m, 1H), 2.55 (m, 1H), 1.65 (m, 3H), 1.29 (s, 9H), 1.02 (d, J = 7.0 Hz, 3H), 0.89 (d, J = 6.2 Hz, 3H), 0.85 (d, J = 6.2 Hz, 3H); ^{13}C NMR (75 MHz, DMSO-d_6) 173.2, 172.2, 171.5, 157.2, 154.6, 129.7, 129.5, 112.9, 77.4, 72.0, 56.0, 54.4, 40.9, 36.6, 27.7, 27.4, 24.1, 22.6, 21.0, 13.8; HRMS (MH^+) calcd for $C_{25}H_{39}N_2O_8$: 495.2706, found: 495.2714.

5(*S*)-[[2(*S*)-[[3-[[2(*R*)-(*tert*-Butoxycarbonylamino)-3-(4-methoxyphenyl)propionyl]amino]-2(*R*)-methylpropionyl]oxy]-4-methylpentanoyl]oxy]-6-oxo-8-phenylocta-2(*E*),7(*E*)-dienoic Acid *tert*-Butyl Ester (16). To a stirred solution of **15** (104 mg, 0.21 mmol) in THF (1.3 mL) were added trichlorobenzoyl chloride (37mL, 0.24 mmol) and triethylamine (37mL, 0.27 mmol). After 3 h, the solvent was evaporated, and a solution of **9** (53 mg, 0.18 mmol) in toluene (2.5 mL) was added to the residue. Triethylamine (30 mL, 0.18 mmol) and DMAP (3 mg, 0.02 mmol) were then added. After 1 h, the reaction mixture was extracted in CH_2Cl_2 and 0.1 N HCl, and the organic phase was washed with brine and dried over $MgSO_4$. After evaporation of the solvents, the residue was purified by flash chromatography (hexane:EtOAc 2:1) to give **16** (110 mg, 85%) as a colorless solid. $[\alpha]_D$ −42.6° (*c* 2.2, $CHCl_3$); IR (neat film) 3350, 2980, 1741, 1713, 1680, 1612, 1514, 1368, 1249, 1171 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 7.74 (d, J = 16.0 Hz, 1H), 7.59 (d, J = 6.1 Hz, 2H), 7.43 (m, 3H), 7.06 (d, J = 8.0 Hz, 2H), 6.85 (d, J = 16.0 Hz, 1H), 6.80 (m, 1H), 6.77 (d, J = 8.0 Hz, 2H), 6.67 (broad s, 1H), 5.87 (d, J = 15.7 Hz, 1H), 5.51 (broad s, 1H), 5.23 (broad d, 1H), 5.14 (dd, J = 4.0, 9.1 Hz, 1H), 4.31 (m, 1H), 3.75 (s, 3H), 3.62 (m, 1H), 3.15 (m, 1H), 3.00 (m, 1H), 2.92 (m, 1H), 2.84 (m, 1H), 2.72 (m, 2H), 1.82 (m, 3H), 1.47 (s, 9H), 1.38 (s, 9H), 1.16 (d, J = 7.0 Hz, 3H), 0.99 (d, J = 6.1 Hz, 3H), 0.96 (d, J = 6.0 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) 192.7, 173.9, 171.5, 170.4, 164.9, 158.2, 155.1, 145.4, 139.7, 133.8, 131.1, 130.1, 128.9, 128.7, 128.6, 126.8, 120.5, 113.7, 80.4, 76.3, 70.6, 55.8, 55.0, 41.6, 40.0, 39.3, 37.9, 33.2, 28.1, 27.9, 24.6, 22.8, 21.4, 14.4; HRMS (MH^+) calcd for $C_{43}H_{59}N_2O_{11}$: 779.4119, found: 779.4114.

(3*S*,6*R*,10*S*,16*S*) 3-Isobutyl-10-(4-methoxybenzyl)-6-methyl-16-(3-phenylacryloyl)-1,4-dioxa-8,11-diazacyclohexadec-13(*E*)-ene-2,5,9,12-tetraone (2). To a stirred solution of compound **16** (106 mg, 0.136 mmol) in CH_2Cl_2 (3.2 mL) at room temperature was added trifluoroacetic acid (1.6 mL). After 45 min, toluene was added and the solvents were evaporated. The residue was dissolved in CH_3CN (14 mL). To the stirred solution at 0 °C were added *O*-benzotriazol-1-yl-*N*,*N*,*N*′,*N*′-bis(pentamethylene)uronium hexafluorophosphate (75 mg, 0.163 mmol), followed by diisopropylethylamine (71 mL, 0.408 mmol). After 30 min, the reaction mixture was extracted in CH_2Cl_2 and 0.1 N HCl. The organic phase was washed with brine and dried over $MgSO_4$. The solvents were evaporated, and the residue was purified by flash chromatography (CH_2Cl_2: acetone 5:1) to give **2** and its 16-epimer (65 mg, 79%) as a 7:1 mixture, respectively. The products were separated by flash chromatography (hexane:acetone 2:1). Compound **2**: $[\alpha]_D$ −8.9° (*c* 1.1, $CHCl_3$); IR (neat film) 3490, 3280, 2960, 1758, 1727, 1678, 1612, 1514, 1248, 1178 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 7.72 (d, J = 16.0 Hz, 1H), 7.55 (m, 2H), 7.44 (m, 3H), 7.14 (d, J = 8.6 Hz, 2H), 7.07 (dt, J = 4.4, 7.1 Hz, 1H), 6.91 (d, J = 16.0 Hz, 1H), 6.85 (d, J = 8.6 Hz, 2H), 6.79 (ddd, J = 4.6, 6.0, 15.1 Hz, 1H), 5.84 (dd, J = 1.3, 15.2 Hz, 1H), 5.72 (dd, J = 2.2, 11.5 Hz, 1H), 5.63 (d, J = 8.0 Hz, 1H), 5.03 (dd, J = 4.7, 9.4 Hz, 1H), 4.82 (bq, J = 7.6 Hz,

1H), 3.80 (s, 3H), 3.43 (m, 2H), 3.20 (dd, J = 5.4, 14.5 Hz, 1H), 3.07 (dd, J = 7.5, 14.5 Hz, 1H), 2.93 (ddd, J = 2.2, 4.6, 14.7 Hz, 1H), 2.75 (m, 1H), 2.55 (m, 1H), 1.80 (m, 2H), 1.60 (m, 1H), 1.27 (d, J = 7.4 Hz, 3H), 0.93 (d, J = 6.5 Hz, 3H), 0.89 (d, J = 6.5 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) 193.6, 175.9, 171.2, 170.1, 165.1, 158.6, 145.6, 140.1, 133.9, 131.3, 130.1, 129.1, 128.6, 128.5, 126.0, 120.4, 114.1, 76.3, 71.3, 55.2, 54.3, 40.4, 39.6, 37.9, 35.3, 34.8, 24.5, 22.7, 21.7, 14.3. Anal. Calcd for $C_{34}H_{40}N_2O_8$: C, 67.53; H, 6.67; N, 4.63. Found: C, 67.02 H, 6.64; N, 4.62. **16-epimer** $[\alpha]_D$ −3.8° (*c* 0.4, $CHCl_3$); IR (neat film) 3300, 2960, 1738, 1680, 1612, 1513, 1248, 1178 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 8.16 (d, J = 16.4 Hz, 1H), 7.72 (m, 2H), 7.43 (m, 3H), 7.16 (d, J = 8.5 Hz, 1H), 7.06 (d, J = 8.6 Hz, 2H), 7.01 (d, J = 16.4 Hz, 1H), 6.75 (m, 2H), 6.68 (d, J = 8.6 Hz, 2H), 6.02 (bd J = 9.7 Hz, 1H), 5.94 (d, J = 15.8 Hz, 1H), 4.99 (dd, J = 5.0, 9.3 Hz, 1H), 4.77 (m, 1H), 3.72 (s, 3H), 3.64 (m, 1H), 3.11 (m, 1H), 2.96 (m, 3H), 2.78 (m, 1H), 2.68 (m, 1H), 1.90 (m, 1H), 1.77 (m, 2H), 1.20 (d, J = 6.9 Hz, 3H), 1.01 (d, J = 6.6 Hz, 3H), 0.98 (d, J = 6.5 Hz, 3H); HRMS (M^+) calcd for $C_{34}H_{40}N_2O_8$: 604.2784, found: 604.2789.

(3*S*,4*S*)-3-(Tetrahydropyran-2-yloxy)-6-phenylhex-5(*E*)-ene-1,4-diol (17). To a stirred solution of **5** (265 mg, 1.23 mmol) in THF (4 mL) at room temperature were added dihydropyran (206 mL, 2.26 mmol) and *p*-toluenesulfonic acid (22 mg, 0.12 mmol). After 3 h, the reaction mixture was worked up in EtOAc and 5% $NaHCO_3$, and the organic phase was washed with brine and dried over $MgSO_4$. The solvents were evaporated, and the residue was dissolved in ether (6 mL). The solution was added to a suspension of LAH (160 mg, 4.22 mmol) in ether (26 mL), and the mixture was stirred at room temperature overnight. Methanol (1 mL) was then added, and the reaction mixture was worked up with EtOAc and 0.1 N HCl. The organic phase was washed with brine and dried over Na_2SO_4. The solvents were then evaporated to give the crude diol **17** (346 mg, 92%). ^{1}H NMR (400 MHz, $CDCl_3$) 7.34 (m, 5H), 6.69 (2d, J = 13.7 Hz, 1H), 6.18 (2 dd, J = 6.4, 13.7 Hz, 1H), 4.60 (2d, J = 7.0, 7.6 Hz, 1H), 4.20 (1m, 1H), 4.10 (1m, 1H), 3.90 (1m, 1H), 3.70 (m, 2H), 3.55 (m, 1H), 1.85 (m, 3H), 1.70 (m, 1H), 1.60 (m, 3H); HRMS (MNH_4^+) calcd for $C_{17}H_{28}NO_4$: 310.2018, found: 310.2025.

(3*S*,4*S*)-2,2-Dimethylpropionic Acid 4-Acetoxy-3-hydroxy-6-phenylhex-5(*E*)-enyl Ester (18). To a stirred solution of compound **17** (320 mg, 1.04 mmol) in CH_2Cl_2 (2 mL) and pyridine (2 mL) at 0 °C were added trimethylacetyl chloride (515 mL, 4.18 mmol) and DMAP (32 mg, 0.20 mmol). After 3 h, acetic anhydride was added and the solution was stirred another 3 h. After workup, the residue was purified by flash chromatography (hexane:EtOAc 4:1) to give 264 mg (59%) of the fully protected compound. The product (208 mg, 0.48 mmol) was dissolved in acetic acid (5 mL) and water (2.5 mL) and stirred at room temperature for 3 h. The reaction mixture was extracted in CH_2Cl_2 and water. The organic phase was washed 5% $NaHCO_3$ and brine and dried over $MgSO_4$. The solvent was evaporated to give **18** (164 mg, 98%): ^{1}H NMR (400 MHz, $CDCl_3$) 7.41 (m, 2H), 7.33 (m, 3H), 6.72 (d, J = 16.0 Hz, 1H), 6.19 (dd, J = 7.6, 16.0 Hz, 1H), 5.38 (m, 1H), 4.34 (m, 1H), 4.22 (m, 1H), 3.85 (m, 1H), 2.38 (broad d, J = 4.2 Hz, 1H), 2.14 (s, 3H), 1.89 (m, 1H), 1.76 (m, 1H), 1.21 (s, 9H).

(3*S*,4*S*)-2,2-Dimethylpropionic Acid 3-Hydroxy-4-methyl-6-phenylhex-5(*E*)-enyl Ester (19). To a stirred suspension of CuI (446 mg, 2.23 mmol) in ether (10 mL) at 0 °C was slowly added a solution of methyllithium in ether (3.1 mL, 1.4 M). After 10 min, **18** (147 mg, 0.42 mmol) in ether (2 mL) was added. After 20 min, the reaction mixture was extracted in CH_2Cl_2 and 0.1 N HCl. The organic phase was washed 5% $NaHCO_3$ and brine and dried over $MgSO_4$. The solvent was evaporated and the residue was purified by flash chromatography (hexane:EtOAc 6:1) to give **19** (44 mg, 34%): ^{1}H NMR (400 MHz, $CDCl_3$) 7.37 (m, 2H), 7.25 (m, 3H), 6.47 (d, J = 16.0 Hz, 1H), 6.17 (dd, J = 8.5, 16.0 Hz, 1H), 4.32 (m, 1H), 4.22 (m, 1H), 3.60 (m, 1H), 2.40 (m, 1H), 2.08 (d, J = 4.0 Hz), 1.90 (m, 1H), 1.74 (m, 1H), 1.22 (s, 9H), 1.17 (d, J = 6.8 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) 178.8, 137.1, 131.5, 131.4, 128.7, 127.3, 126.1, 72.1, 61.8, 43.6, 38.7, 33.6, 27.2, 16.7.

(5*S*,6*R*)-5-Hydroxy-6-methyl-8-phenylocta-2(*E*),7(*E*)-dienoic Acid *tert*-Butyl Ester (20). To a stirred solution of **19** (44 mg, 0.145 mmol) in THF (1.5 mL) at room temperature were added dihydropyran (80 mL, 0.86 mmol) and PTSA (3 mg, 0.02 mmol). After 2 h, the reaction mixture was worked up in CH_2Cl_2 and 5% $NaHCO_3$. The organic phase was washed brine and dried over $MgSO_4$. The solvent was evaporated, and the residue was dissolved in THF (1.5 mL). To the stirred solution at 0°C was added LAH (6.2 mg, 0.16 mmol). After 15 min, the reaction mixture was worked up in CH_2Cl_2 and 0.1 N HCl. The organic phase was washed $NaHCO_3$ 5% and brine and dried over $MgSO_4$. After evaporation of the solvents, the residue was dissolved in CH_2Cl_2 (0.5 mL) and added to a stirred solution of DMSO (72 mL, 0.52 mmol) and oxalyl chloride (24 mL, 0.34 mmol) in CH_2Cl_2 (2 mL) at −78 °C. After 30 min, triethylamine (72 mL, 0.52 mmol) was added and the solution was warmed up to 0 °C. After 15 min, (*tert*-butoxycarbonylmethylene)triphenylphosphorane (64 mg, 0.17 mmol) was added and the solution was stirred for another 20 min. The mixture was then worked up in CH_2Cl_2 and 0.1 N HCl. The organic phase was washed 5% $NaHCO_3$ and brine and dried over $MgSO_4$. After evaporation of the solvent, the residue was purified by flash chromatography (hexane:EtOAc 5:1) to give 38 mg, (65%) from **19**. The product was then dissolved in acetic acid (1.0 mL) and water (0.5 mL), and the solution was stirred at 40°C for 30 min. The reaction mixture was extracted in CH_2Cl_2 and water. The organic phase was washed 5% $NaHCO_3$ and brine and dried over $MgSO_4$. The solvent was evaporated and the residue purified by flash chromatography (hexane:EtOAc 5:1) to give **20** (24 mg, 80%). $[\alpha]_D$ +46.7° (*c* 0.83, $CHCl_3$); IR (neat film) 3450, 2930, 1700, 1654, 1368, 1154 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 7.38 (m, 2H), 7.32 (m, 2H), 7.24 (m, 1H), 6.92 (dt, J = 7.3, 15.6 Hz, 1H), 6.49 (d, J = 16.0 Hz, 1H), 6.15 (dd, J = 8.6, 16.0 Hz, 1H), 5.86 (dt, J = 1.4, 15.6 Hz, 1H), 3.66 (m, 1H), 2.44 (m, 2H), 2.36 (m, 1H), 1.76 (d, J = 3.9 Hz, 1H), 1.50 (s, 9H), 1.16 (d, J = 6.9 Hz, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) 165.7, 144.0, 137.0, 131.9, 131.0, 128.5, 127.4, 126.2, 125.5, 80.2, 73.9, 43.2, 37.2, 28.1, 16.8); HRMS (MNH_4^+) calcd for $C_{19}H_{30}NO_3$: 320.2226, found: 320.2219.

2(*S*)-[[3-[[2(*R*)-(*tert*-Butoxycarbonylamino)-3-(3-chloro-4-methoxyphenyl)propionyl]amino]-2(*R*)-methylpropionyl]oxy]-4-methylpentanoic Acid (24). A mixture of **15** (600 mg, 1.21 mmol) and freshly distilled sulfuryl chloride (0.107 mL, 1.33 mmol) in glacial acetic acid (6 mL) was heated at 55 °C for 30 min. The reaction mixture was then cooled down to room temperature, and the solvent was evaporated to give 718 mg of the deprotected chloro tyrosine derivative. This crude product was then dissolved in CH_2Cl_2 (14 mL) and BOC-ON (449 mg, 1.82 mmol) and triethylamine (372 mL, 2.69 mmol) were added. After stirring at room temperature overnight, 0.1 N HCl was added and the reaction mixture extracted with CH_2Cl_2. The organic phase was washed with water and brine and dried over $MgSO_4$. The solvent was evaporated, and the residue was purified by flash chromatography (from $CHCl_3$:EtOAc 95:5 to $CHCl_3$:EtOAc:MeOH 85:10:5) to give 438 mg (68%) of **24** as a light yellow solid: mp 75−80 °C; $[\alpha]_D$ −21.8° (*c* 5.55, MeOH); IR (neat film) 3305, 2960, 1733, 1658, 1590, 1504, 1260 cm^{-1}; ^{1}H NMR (400 MHz, DMSO-d_6) 8.60 (broad s, 1H,), 7.29 (s, 1H), 7.16 (d, J = 8.4 Hz, 1H), 7.02 (d, J = 8.4 Hz, 1H), 6.91 (d, J = 8.8 Hz, 1H), 4.81 (m, 1H), 4.05 (m, 1H), 3.80 (s, 3H), 3.27 (m, 2H), 2.81 (m, 1H), 2.65 (m, 1H), 2.57 (m, 1H), 1.65 (m, 3H), 1.29(s, 9H), 1.03 (d, J = 7.0 Hz, 3H), 0.88 (d, J = 6.2 Hz, 3H), 0.85 (d, J = 6.2 Hz, 3H); ^{13}C NMR (400 MHz, DMSO-d_6) 173.3, 171.3, 154.7, 152.5, 130.9, 130.0, 128.6, 128.4, 127.7, 124.8, 119.9, 111.9, 77.5, 71.3, 55.6, 55.5, 40.8, 36.1, 27.6, 24.0, 22.5, 21.0, 13.8; HRMS (MH^+) calcd for $C_{25}H_{38}ClN_2O_8$: 529.2317, found: 529.2302.

(3*S*,6*R*,10*S*,16*S*)-3-Isobutyl-10-(3-chloro-4-methoxybenzyl)-6-methyl-16-(3-phenylacryloyl)-1,4-dioxa-8,11-diazacyclohexadec-13(*E*)-ene-2,5,9,12-tetraone (1). **9** and **24** were coupled with the same procedure used for the preparation of **16** and gave the coupling product in 79% (64 mg): $[\alpha]_D$ −41.2° (*c* 2.0, $CHCl_3$); IR (neat film) 3350, 3000, 1741, 1713, 1685, 1662, 1610, 1504, 1258, 1170 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 7.74 (d, J = 16.0 Hz, 1H), 7.58 (d, J = 6.6 Hz, 1H),

7.42 (s, 1H), 7.02 (d, J = 7.5 Hz, 1H), 6.82 (m, 2H), 6.79 (d, J = 8.3 Hz, 1H), 5.89 (d, J = 15.6 Hz, 1H), 5.54 (broad s, 1H), 5.32 (broad s, 1H), 5.16 (dd, J = 4.0, 9.1 Hz, 1H), 4.34 (m, 1H), 3.86 (s, 3H), 3.68 (m, 1H), 3.17 (m, 1H), 3.01 (m, 1H), 2.86 (m, 2H), 2.73 (m, 2H), 1.83 (m, 3H), 1.60 (m, 1H), 1.49 (s, 9H), 1.39 (s, 9H), 1.19 (d, J = 7.0 Hz, 3H), 1.00 (d, J = 5.9 Hz, 3H), 0.97 (d, J = 6.0 Hz, 3H); ^{13}C NMR (400 MHz, $CDCl_3$) 193.6, 173.9, 171.3, 170.9, 165.1, 155.2, 153.7, 145.6, 139.8, 133.9, 131.2, 131.0, 130.1, 129.0, 128.7, 128.5, 127.0, 121.9, 120.6, 112.1, 80.6, 79.7, 76.4, 70.7, 56.0, 55.7, 41.8, 40.3, 39.5, 37.7, 33.4, 28.2, 28.0, 24.8, 23.0, 21.5, 14.5; HRMS (M^+) calcd for $C_{43}H_{58}ClN_2O_{11}$: 813.3729, found: 813.3735. The deprotection and the cyclization steps were run as compound **2** (31 mg, 64%): $[\alpha]_D$ −10.0° (*c* 1.07, $CHCl_3$); IR (neat film) 3405, 3280, 3000, 1758, 1725, 1674, 1610, 1504, 1172 cm^{-1}; ^{1}H NMR (400 MHz, $CDCl_3$) 7.70 (d, J = 16.0 Hz, 1H), 7.55 (m, 1H), 7.40 (m, 3H), 7.24 (d, J = 2.0 Hz, 1H), 6.92 (d, J = 16.0 Hz, 1H), 6.85 (d, J = 8.4 Hz, 1H), 6.77 (ddd, J = 4.7, 10.5, 15.2 Hz, 1H), 5.98 (d, J = 8.3 Hz, 1H), 5.89 (d, J = 15.2 Hz, 1H), 5.69 (dd, J = 2.1, 11.5 Hz, 1H), 5.02 (dd, J = 4.7, 9.5 Hz, 1H), 4.82 (m, 1H), 3.87 (s, 3H), 3.47 (m, 1H), 3.41 (m, 1H), 3.18 (dd, J = 5.2, 14.5 Hz, 1H), 2.99 (dd, J = 8.0, 14.5 Hz, 1H), 2.93 (dd, J = 2.4, 12.4 Hz, 1H), 2.74 (m, 1H), 2.53 (m, 1H), 1.77 (m, 2H), 1.58 (m, 1H), 1.26 (d, J = 7.2 Hz, 3H), 0.91 (d, J = 6.5 Hz, 3H), 0.87 (d, J = 6.4 Hz, 3H); ^{13}C NMR (400 MHz, $CDCl_3$) 193.5, 175.6, 171.0, 170.1, 165.3, 153.9, 145.6, 140.2, 133.9, 131.3, 130.9, 129.8, 129.1, 128.6, 128.3, 126.0, 122.4, 120.4, 112.2, 76.3, 71.3, 56.1, 54.1, 40.7, 39.6, 37.9, 35.0, 34.8, 24.6, 22.8, 21.7, 14.2; IRMS (M^+) calcd for $C_{34}H_{39}ClN_2O_8$: 638.2395, found: 638.2388.

Cryptophycin C. Cryptophycin C was prepared by the coupling between **20** and **24** followed by the deprotection and the cyclization steps (37 mg, 58% overall). ^{1}H NMR, ^{13}C NMR, IR, HRMS, and $[\alpha]_D$ are in perfect agreement with the reported values.[4]

Cryptophycin D. Cryptophycin D was prepared by the coupling between **15** and **20** followed by the deprotection and the cyclization steps (7 mg, 35% overall). ^{1}H NMR, ^{13}C NMR, IR, HRMS, and $[\alpha]_D$ are in perfect agreement with the reported values.[4]

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Supporting Information Available: Copies of ^{1}H and/or ^{13}C NMR spectra for **1**, **2**, **4**−**9**, **11, 12**, **14**−**20**, **24** and Cryptophycins A, C, D (36 pages). This material is contained in libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.

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