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Total Synthesis of a Macrocyclic Antibiotic, Micrococcin P_1

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The first total synthesis of a macrocyclic antibiotic, micrococcin P_1 (**1**), was achieved. This antibiotic has a unique structure and is constructed of four components called Fragments A, B, C, and D. In particular, the structures of the central pyridine skeleton (Fragment A) and the exocyclic side-chain (Fragment D) of **1** are slightly different from those of a similar antibiotic, micrococcin P (**2**). By various chemical modifications of the synthetic method for **2**, the synthesis of the central 2,3,6-tris(substituted thiazolyl)pyridine segment [Fragment A—C] **15** from ethyl 2-[6-dimethoxymethyl-2-(1-ethoxyvinyl)-3-pyridyl]thiazole-4-carboxylate (**9**), followed by coupling of **15** with the Fragments B and D moieties, synthesized independently, gave the protected Fragment A-B-C-D segment. Final intramolecular cyclization by using (benzotriazol-1-yloxy)tris(dimethylamino)phosphonium hexafluorophosphate as a condensing agent under high-dilution conditions and then deprotection of all the protecting groups with trifluoroacetic acid were effected successfully to give the expected synthetic **1**.

Micrococcin P_1 (**1**),[1] isolated from the culture of *Bacillus pumilus*, is a very interesting macrocyclic antibiotic. So far, many structurally similar antibiotics, such as GE 2270 A[2] and thiocillins I—III,[3] have been also isolated from various kinds of strains. However, no total synthesis of any similar antibiotic has yet been reported except for the recent synthesis of micrococcin P (**2**).[4,5] Micrococcin P_1 (**1**), like **2**,[6] features a very unique structure. It includes commonly a characteristic main central structure, a central 2,3,6-tris-(substituted thiazolyl)pyridine skeleton called Fragment A-C **15** (thiazole; Tz, pyridine; Py). Moreover, interestingly, the two sites in the structures of the respective Fragments A and D of **1** and **2** are reversed, as shown in Fig. 1. That is, the P_1 **1** is constructed of a (1*S*,2*R*)-1-amino-1-[4-(2-pyridyl)thiazol-2-yl]-2-propanol segment and a (*Z*)-2-amino-2-butenoic acid (ΔAbu) residue in Fragments A and D, whereas the P **2** includes a (*Z*)-1-[4-(2-pyridyl)thiazol-2-yl]-1-propenylamine segment and L-threonine residue, respectively. Therefore, a different methodology for the synthesis of **1** has to be devised. Although the configurational structure of **1** has been determined by the ^1H and ^{13}C NMR spectral data, the chemical and physical properties, unfortunately, have not been reported. The interesting structure as well as the bioactivity of **1**, which exhibits inhibitory action of bacterial protein synthesis similar to that of **2**, attracted us and prompted us to investigate the synthesis, structural confirmation, and structure-bioactivity relationship.

More recently,[5,7] we have reported briefly the total synthesis of **1** by the coupling of a very promising key compound, Fragment A-C (**15**),[4,7] with the protected Fragments B and D derivatives[4,8] and then final cyclization of the obtained Fragment A-B-C-D segment by using BOP[9] as a condensing agent under high-dilution conditions.

In this paper, having taken advantage of the synthetic method for **2**, we report the first total synthesis of **1** as well. We describe in detail a convenient synthesis of **1** from the

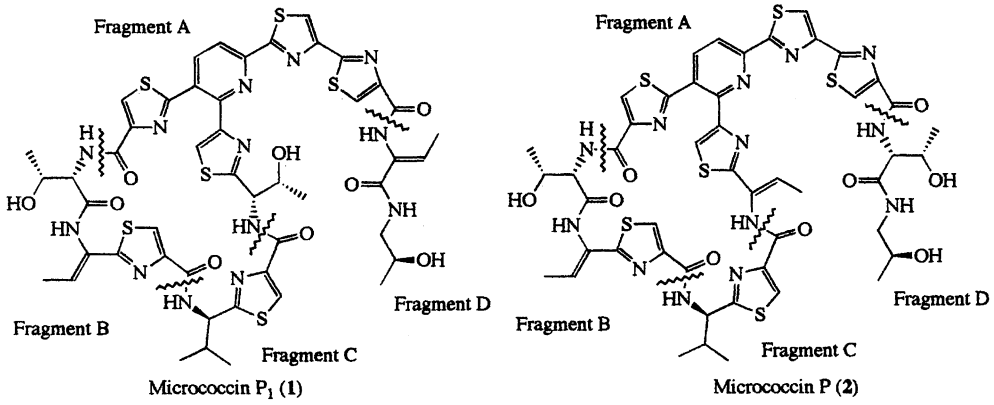


Fig. 1.

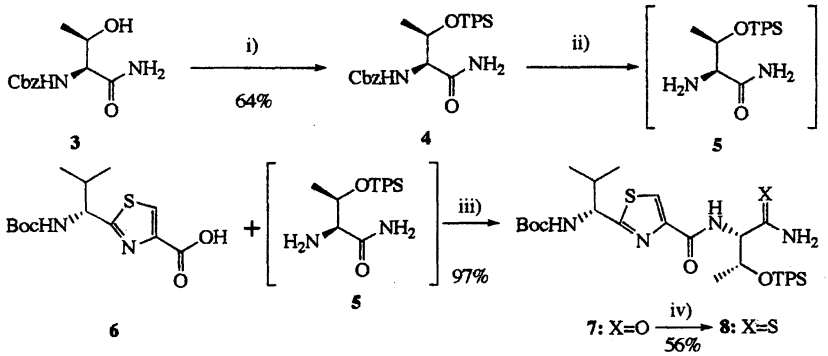
newly-synthesized Fragments A, B, C, and D segments. Furthermore, the configurational structure of the natural **1** could be confirmed by the agreement of the ^1H and ^{13}C NMR spectral analyses as well as the determination of the chemical and physical properties (Mp, $[\alpha]_D$, IR, and UV), which are very similar to those of **2**.

Results and Discussion

First of all, to synthesize the Fragment A-C segment **15** of **1**, differently from the case of the synthesis of **2**, the *t*-butyldiphenylsilyl (TPS) group was chosen as the *O*-protecting group so as not to dehydrate the hydroxy group of (1*S*,2*R*)-1-amino-1-[4-(2-pyridyl)thiazol-2-yl]-2-propanol segment in Fragment A. That is, the two secondary hydroxy groups in the precursor of Fragment A-C-D segment **21** were later subjected to different protection, because it was found that, if the protecting groups are the same, simultaneous dehydration of the two sites readily took place. Accordingly, the hydroxy group of *N*-benzyloxycarbonyl (Cbz)-L-Thr-NH_2 (**3**) was protected with TPSCl in the presence of imidazole to give Cbz-L-Thr(TPS)-NH_2 (**4**), the Cbz group of which was then deprotected by hydrogenolysis with 10% Pd–C in EtOH to give H-L-Thr(TPS)-NH_2 (**5**). Subsequently, without isolation, the formed **5** as an amine (*N*-) component was subjected to coupling with 2-[(*R*)-1-(*t*-butoxycarbonylamino)-2-methylpropyl]thiazole-4-carboxylic acid (**6**: Fragment C)[10,11] as a carboxy (*C*-) component. Coupling between **5** and **6** using diphenoxyphosphinoyl azide (DPPA)[12] as a condensing agent in the presence of Et_3N in DMF was carried out to give the corresponding *N*-(4-thiazolecarbonyl)threonine derivative **7**. Furthermore, the compound **7** was thioamidated with Lawesson's reagent[13] in 1,2-dimethoxyethane (DME) to give the expected protected *N*-(4-thiazolecarbonyl)threonine thioamide derivative **8** as the precursor for the synthesis of the desired Fragment A-C, as shown in Scheme 1.

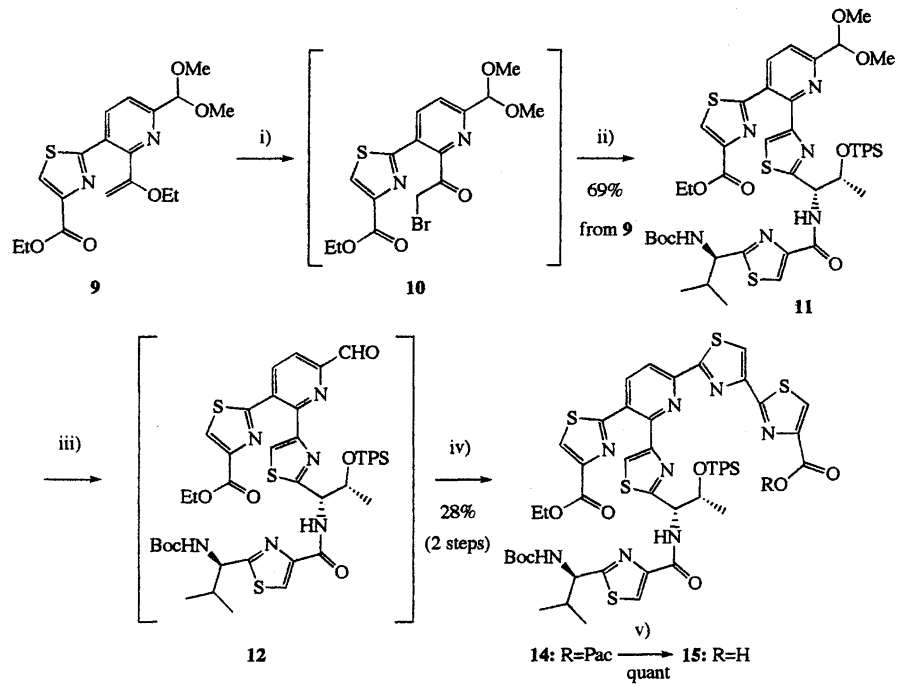
Subsequently, compound **8**, thus obtained, was submitted to the thiazole ring formation of Fragment A. The thiazolation of the bromoacetyl group of ethyl 2-(2-bromoacetyl-6-dimethoxymethylpyridin-3-yl)thiazole-4-carboxylate (**10**),[5,7] derived by the reaction of ethyl 2-[6-dimethoxymethyl-2-(1-ethoxyvinyl)-3-pyridyl]thiazole-4-carboxylate (**9**) with *N*-bromosuccinimide (NBS) and water, was performed as follows. Similarly to the case of the synthesis of the Fragment A-C of **2**,[5] direct cyclization of **10** with **8** by using successively $KHCO_3$, trifluoroacetic anhydride (TFAA), and 28% aqueous NH_3 was carried out to give the corresponding 6-dimethoxymethyl-2,3-bis(substituted thiazolyl)-pyridine derivative **11**[14] in 69% yield in four steps from **9**. Furthermore, the formylation of the dimethoxymethyl group of **11** with 2 M (1 M = 1 mol dm^{-3}) HCl in THF was performed to give the corresponding 6-formylpyridine derivative **12**, the formyl group of which was then in situ transformed to a thiazole ring. That is, firstly, the thiazolation of the formyl group with phenacyl (Pac) (*R*)-2-[(3-*t*-butoxycarbonyl-2,2-dimethyl)thiazolidin-4-yl]thiazole-4-carboxylate (**13**)[7] using trifluoroacetic acid (TFA), followed by oxidation with MnO_2 in toluene gave the corresponding 2′-(2-pyridyl)-2,4′-bithiazole-4-carboxylate derivative **14**, according to the Shioiri method.[11] Finally, the Pac ester **14** was hydrolyzed with 1 M LiOH to give the corresponding 2,4′-bithiazole-4-carboxylic acid derivative **15** almost quantitatively, as shown in Scheme 2. As the result, the convenient synthesis of the main Fragment A-C derivative, which is a very essential intermediate for the synthesis of **1**, was achieved.

On the other hand, to synthesize the side chain Fragment D moiety, firstly, the protection of the hydroxy group of the starting Boc-L-Thr-OBn (**16**) with *t*-butyldimethylsilyl chloride (TBSCl) in the presence of imidazole gave the corresponding Thr(TBS) derivative, the benzyl (Bn) ester of which was in situ hydrogenolyzed with 10% Pd–C in EtOH to give Boc-L-Thr (TBS)-OH (**17**). Secondly, coupling of **17** with *S*-(+)-1-amino-2-propanol using BOP gave (2*S*,3*R*)-2-(*t*-butoxycarbonyl)amino-3-(*t*-butyldimethylsiloxy)-*N*-[(*S*)-2-hydroxypropyl]butanamide (**18**), the hydroxy group of which was then acetylated with acetic anhydride (Ac_2O) in the presence of pyridine to give the corresponding *N*-[(*S*)-2-acetoxypropyl)butanamide derivative **19** almost quantitatively. Lastly, deprotection of the Boc group of **19** with TFA in the presence of MS4A (molecular sieves) proceeded smoothly to give the expected (1*S*,2*R*)-2-amino-3-(*t*-butyldimethylsiloxy)-*N*-[(*S*)-2-acetoxypropyl]butanamide (**20**) as the precursor of Fragment D segment, as shown in Scheme 3.



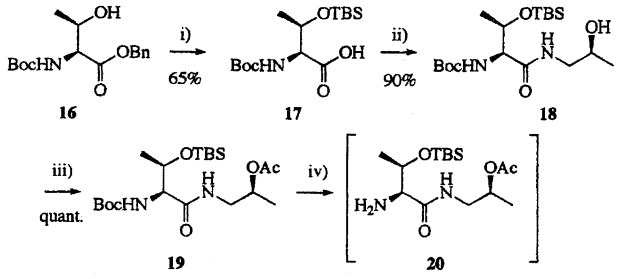
i) TPSCl, imidazole, DMF, 0 °C, 30 min, r.t., 24 h, ii) 10% Pd-C, H_2, EtOH, r.t., 3 h, iii) DPPA, Et_3N, DMF, 0 °C, 3 h, r.t., overnight, iv) Lawesson's reagent, DME, r.t., 12 h.

Scheme 1.



i) NBS, THF / H_2O, r.t., 5 min, ii) a) **8**, $KHCO_3$, 0 °C, 30 min, r.t., overnight, b) TFAA, pyridine, 0 °C, 2 h, c) 28% aqueous NH_3, ethyl acetate, 0 °C, 15 min, iii) 2M HCl-THF, r.t., 12 h, iv) a) **13**, TFA, CH_2Cl_2, r.t., 1 h, b) toluene, r.t., 15 min, b) MnO_2, toluene, overnight, v) 1M LiOH, THF, 0 °C, 2 h.

Scheme 2.

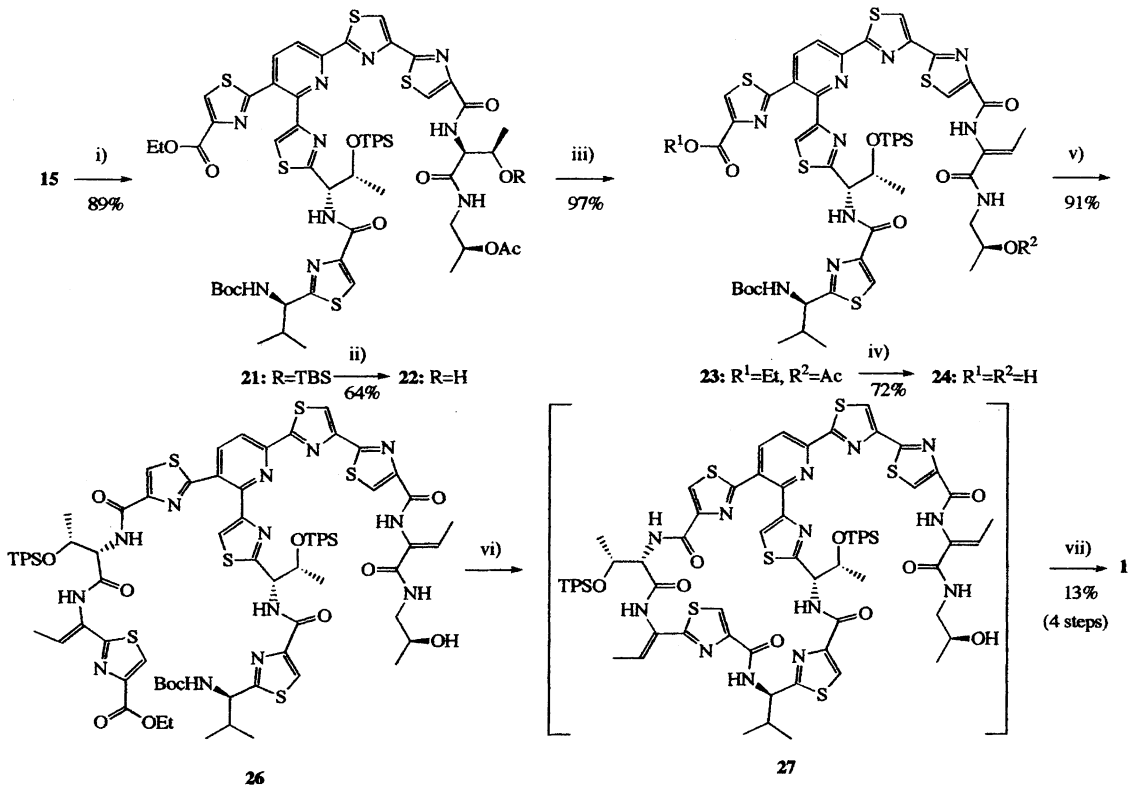


i) a) TBSCl, imidazole, CH_2Cl_2, 0 °C, 30 min, r.t., overnight, b) 10%Pd-C, H_2, EtOH, r.t., 3 h, ii) (*S*)-(+)-1-amino-2-propanol, BOP, (*i*-Pr)$_2$NEt, CH_3CN, 0 °C, 30 min, r.t., overnight, iii) Ac_2O, pyridine, r.t., 2 h, iv) TFA / CH_2Cl_2, r.t., 1 h, MS4A.

Scheme 3.

Finally, similarly to the last stage in the synthesis of **2**,[5] firstly, the compound **20** prepared above was subjected to coupling with the 4-carboxy group of the Fragment A-C skeleton **15**. The fragment condensation of the *C*-component **15** with **20** as the *N*-component by the BOP method was carried out to give the precursor of the protected Fragment A-C-D segment **21**. Secondly, selective deprotection of only the TBS protecting group of Thr residue in **21** with 70% AcOH, followed by mesylation of the deprotected intermediate **22** with methanesulfonyl (mesyl) chloride (MsCl) in the presence of Et_3N and then β-elimination with 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) gave the protected Fragment A-C-D derivative **23** in 97% yield. The geometric structure of the formed ΔAbu residue was unambiguously determined to have (*Z*)-geometry by comparison with the chemical shifts of the 3-methyl and olefinic protons of the (*E*)- and (*Z*)-ΔAbu derivatives prepared independently by the condensation of alkyl 2-oxoalkanoate with an appropriate haloacetamide.[15] Then, further hydrolysis of the ethyl ester and then deprotection of the acetyl group of **23** with 1M LiOH was performed to give the corresponding 2-(3-pyridyl)thiazole-4-carboxylic acid derivative **24**. Moreover, fragment condensation of **24** as the *C*-component with the *N*-component ethyl 2-{(*Z*)-1-[(2*S*,3*R*)-2-amino-3-(*t*-butyldiphenylsiloxy)butanoylamino]-1-propenyl}thiazole-4-carboxylate (**25**)[5] synthesized previously by the BOP method gave the protected Fragment A-B-C-D derivative (**26**) in 91% yield. Lastly, hydrolysis of the ethyl ester **26** with 1 M LiOH and then deprotection of the Boc group with a mixture of TFA and CH_2Cl_2 (4 : 6 v/v), followed by intramolecular cyclization using BOP in the presence of (*i*-Pr)$_2$NEt in DMF under high-dilution conditions at 0 °C for 1 h and then at room temperature overnight gave the protected micrococcin P_1 derivative **27** as a colorless crude syrup. Then, without pure isolation, careful deprotection of the TPS group with tetrabutylammonium fluoride (TBFA) was carried out to give the expected micrococcin P_1 (**1**) as a reddish syrup, as shown in Scheme 4. The thus-obtained syrup was purified on a TLC using a mixture of $CHCl_3$ and MeOH (20 : 1 v/v) as the eluent to give crude crystals. Recrystallization from MeOH–EtOAc gave **1** as colorless crystals in 13% yield in four steps from **26**.

The chemical and physical constants of the synthetic **1** {mp 215—245 °C, $[\alpha]_D^{24}$ +40.0° (*c* 0.50, 90% EtOH), λ_{max} 324.6 nm} were first obtained. Furthermore, it was found that the ^1H and ^{13}C NMR (in DMSO-d_6) spectral data of the synthetic **1** were identical with all of those of the natural **1**. Accordingly, the configurational structure of **1** could be clearly confirmed by the identification of the physical properties (NMR) as well as by satisfactory chemical analysis. In particular, from the ^1H NMR spectrum of synthetic **1**, the appearance of the chemical shifts of the 4,5-vicinal protons



i) **20**, BOP, $(i\text{-Pr})_2$NEt, DMF, 0 °C, 30 min, r.t., overnight, ii) 70%AcOH-THF, r.t., 72 h, iii) a) MsCl, Et_3N, CH_2Cl_2, 0 °C, 10 min., b) DBU, CH_2Cl_2, 0 °C, 30 min, r.t., overnight, iv) 1M LiOH, THF, 0 °C, 3 h, v) **25**, BOP, $(i\text{-Pr})_2$NEt, DMF, 0 °C, 30 min, r.t., overnight, vi) (a) 1M LiOH, THF, 0 °C, 1 h, r.t., 6 h, (b) TFA:CH_2Cl_2=40:60, r.t., 3 h, MS4A, (c) BOP, $(i\text{-Pr})_2$NEt, DMF, 0 °C, 1 h, r.t., overnight, vii) TBAF, THF, r.t., 30 min.

Scheme 4.

of the pyridine ring at δ = 8.17 and 8.34 (d, 2H, J = 7.0 Hz) as doublets and the six thiazole ring protons at δ = 7.78, 7.98, 8.08, 8.25, 8.29, 8.41 as singlets supports the formation of **1**, in addition to the agreement of the ^{13}C NMR spectral data.

In conclusion, the first total synthesis of **1** was accomplished by the effective selection of the protecting groups, deprotecting reagents, and the cyclization as well as the useful synthesis of the promising 2,3,6-trisubstituted pyridine skeleton. Furthermore, the synthetic methodology developed here was found to be applicable effectively to the synthesis of similar macrocyclic antibiotics, such as GE 2270 A,[2] thiocillins,[3] and amythiamicin.[16]

Experimental

The melting points were measured using a Yamato (Model Mp-21) micromelting point apparatus, and are uncorrected. The IR spectra were recorded using a Hitachi EPI-G2 spectrometer in KBr. The ^{1}H NMR and ^{13}C NMR spectra were measured with JEOL EX 90, FX 200, and JNE 500 spectrometer in $CDCl_3$, DMSO-d_6, C_6D_6, or CD_3OD solution with tetramethylsilane used as the internal standard. The specific rotations were measured in a 0.5 dm tube using a JASCO DIP-4 polarimeter in MeOH or H_2O. Thin-layer chromatography (TLC) was done with Merck silica-gel 60Art 5554 plates and column chromatography was carried out with Merck silica-gel 60 or Wakogel C-300. High-pressure liquid chromatography (HPLC) analyses and separations were done on the following columns using a mixture of MeOH and $CHCl_3$ (5 : 95 v/v) with a flow rate of 2.0 ml min^{-1} by detecting UV (254 nm) absorption: TSK gel Silica-60 (7.8 mmIDE×60 cmL).

Starting Materials. L-Thr-OH, L-Cys-OH, and D-Val-OH, (*S*)-(+)-1-amino-2-propanol were purchased from Nippon Rikagakuyakuhin Co., Ltd., Merck KGaA Co., Ltd., and Sigma-Aldrich Japan K. K., respectively.

(2*S*, 3*R*)-2-Benzyloxycarbonylamino-3-(*t*-butyldiphenylsiloxy)butanamide (4). A solution of **3** (18.94 g, 75.10 mmol) and *t*-butyldiphenylchlorosilane (21.22 ml, 82.60 mmol) in the presence of imidazole (12.78 g, 188.0 mmol) in DMF (200 ml) was stirred at 0 °C for 1 h and then overnight at room temperature. The reaction mixture was diluted with water (150 ml) and extracted with EtOAc (3×80 ml). The combined extracts were washed with brine (3×50 ml) and dried over anhydrous Na_2SO_4. Concentration in vacuo gave a syrup, which was purified on a silica-gel column using a mixture of hexane and EtOAc (1 : 1 v/v) to give **4** as a colorless syrup. Yield 23.7 g (64%). $[\alpha]_D^{24}$ −2.4° (*c* 1.0, MeOH). IR 3430, 3358, 2932, 2854, 1683 cm^{-1}. ^{1}H NMR δ = 0.86 (d, 3H, Thr's CH_3, J = 6.3 Hz), 0.96 (s, 9H, TPS's CH_3×3), 4.02—4.07 (m, 1H, Thr's β-H), 4.23—4.27 (m, 1H, Thr's α-H), 5.08 (s, 2H, Cbz's CH_2), 6.89 (br d, 1H, CHN*H*, J = 9.6 Hz), 7.28—7.67 (m, 17H, TPS's Ph×2, Cbz's Ph, and NH_2). Found: C, 67.75; H, 7.18; N, 5.21%. Calcd for $C_{28}H_{34}N_2O_4Si$: C, 67.99; H, 6.93; N, 5.66%.

(2*S*, 3*R*)-2-{2-[(*R*)-1-(*t*-Butoxycarbonylamino)-2-methylpropyl]thiazol-4-ylcarbonylamino}-3-(*t*-butyldiphenylsiloxy)propanamide (7). A suspension of **4** (10.64 g, 21.70 mmol) and 10% Pd–C (1.1 g) in EtOH (150 ml) was stirred under H_2 gas stream for 4 h at room temperature. The 10% Pd–C was filtered off and the filtrate was concentrated in vacuo to give a colorless syrup **5**, which was dissolved in DMF (100 ml). To the resulting solution was added **6** (5.00 g, 16.40 mmol) and DPPA (3.92 ml, 18.10

mmol) in the presence of Et_3N (2.51 ml, 18.10 mmol) under cooling. After the mixture was stirred for 1 h at 0 °C and then overnight at room temperature, the reaction mixture was diluted with water (100 ml). The resulting solution was extracted with EtOAc (3×80 ml) and the combined extracts were washed with brine (3×50 ml), 10% citric acid (2×50 ml), and saturated aqueous $NaHCO_3$ solution (2×50 ml), and then dried over anhydrous Na_2SO_4. Concentration in vacuo gave a brownish syrup, which was purified on a silica-gel column using a mixture of hexane and EtOAc (1 : 1 v/v) to give crystals. Recrystallization from hexane–EtOAc gave **7** as colorless crystals. Yield 97% (10.28 g). Mp 110.0—111.5 °C. $[\alpha]_D^{24}$ −30.8° (*c* 0.59, MeOH). IR 3778, 3376, 2962, 2254, 1680 cm^{-1}. ^1H NMR δ = 0.85 and 0.86 (each d, 6H, $CH(CH_3)_2$, J = 6.7 Hz), 0.89 (d, 3H, Thr's CH_3, J = 6.7 Hz), 0.97 (s, 9H, TPS's CH_3×3), 1.40 (s, 9H, Boc), 2.23—2.27 (m, 1H, $CH(CH_3)_2$), 4.38—4.42 (m, 2H, Thr's α-H and β-H), 4.64—4.67 (m, 1H, CHNHBoc), 7.35—7.63 (m, 12H, TPS's Ph×2 and NH_2), 7.74 (br d, 1H, CHN*H*, J = 8.2 Hz), 8.10 (br d, 1H, CHN*H*, J = 9.2 Hz), 8.20 (s, 1H, Tz's H). Found: C, 61.86; H, 7.64; N, 8.30%. Calcd for $C_{33}H_{46}N_4O_5SSi$: C, 62.04; H, 7.26; N, 8.77%.

(2*S*,3*R*)-2-{2-[(*R*)-1-(*t*-Butoxycarbonylamino)-2-methylpropyl]thiazol-4-ylcarbonylamino}-3-(*t*-butyldiphenylsiloxy)propanethioamide (8). A solution of **7** (9.92 g, 15.43 mmol) and Lawesson's reagent (3.43 g, 8.49 mmol) in DME (250 ml) was stirred at room temperature for 6 h. The reaction mixture was concentrated in vacuo to give a green syrup, which was purified on a silica-gel column using a mixture of hexane and EtOAc to give a solid material. Recrystallization from a hexane–EtOAc gave **8** as colorless crystals. Yield 56% (5.72 g). Mp 95—96 °C. $[\alpha]_D^{24}$ −26.6° (*c* 0.58, MeOH). IR 3400, 2926, 2230, 1896, 1716 cm^{-1}. ^1H NMR δ = 0.87 and 0.88 (each d, 6H, $CH(CH_3)_2$, J = 6.4 Hz), 0.90 (d, 3H, Thr's CH_3, J = 6.7 Hz), 0.95 (s, 9H, TPS's CH_3×3), 1.40 (s, 9H, Boc), 2.25—2.28 (m, 1H, $CH(CH_3)_2$), 4.40—4.60 (m, 1H, Thr's β-H), 4.67—4.70 (m, 2H, Thr's α-H and CHNHBoc), 7.36—7.64 (m, 10H, TPS's Ph×2), 7.74 (br d, 1H, CHN*H*, J = 8.2 Hz), 8.21 (s, 1H, Tz's H), 8.25 (br d, 1H, CHN*H*, J = 9.1 Hz), 9.52 (br s, 1H, NH), 9.85 (br s, 1H, NH). Found: C, 59.72; H, 7.31; N, 8.49%. Calcd for $C_{33}H_{46}N_4O_4S_2Si\cdot 1/2H_2O$: C, 59.70; H, 7.14; N, 8.44%.

Ethyl 2-(2-{2-[(1*S*,2*R*)-1-{2-[(*R*)-1-(*t*-Butoxycarbonylamino)-2-methylpropyl]thiazol-4-ylcarbonylamino}-2-(*t*-butyldiphenylsiloxy)propyl]thiazol-4-yl}-6-dimethoxymethylpyridin-3-yl)-thiazole-4-carboxylate (11). To a solution of **9** (2.17 g, 5.74 mmol) in a mixture of THF and H_2O (50 ml, 1 : 1) were added, with stirring, $CaCO_3$ (600 mg, 6.49 mmol) and NBS (1.13 g, 6.32 mmol) at room temperature. After the mixture was stirred for 5 min, the reaction mixture was extracted with diethyl ether (2×20 ml) and the combined extracts were washed with H_2O (2×10 ml) and then dried over anhydrous Na_2SO_4. Concentration in vacuo under 10 °C gave **10** as a colorless syrup. A solution of **8** (4.60 g, 6.98 mmol) in DME (100 ml) and $KHCO_3$ (4.60 g, 46.00 mmol) was stirred under cooling for 5 min and then added to a solution of the above obtained **10**. After the mixture was stirred under cooling for 30 min and then overnight at room temperature, the reaction mixture was concentrated in vacuo to give a brownish syrup. A solution of the syrup in $CHCl_3$ (150 ml) was washed with brine (50 ml) and then concentrated in vacuo to give a syrup, which was further dissolved in DME (100 ml). After the solution was cooled, TFAA (2.21 ml, 15.98 mmol) and pyridine (3.22 ml, 39.95 mmol) were added to the resultant solution, which was stirred for 2 h and then was made alkaline to pH 7—9 with Et_3N. Concentration in vacuo gave a brownish syrup, which was dissolved in EtOAc (100 ml) and then washed with brine (2×20 ml). The resultant solution was stirred with 28% aqueous NH_3 (1.69 ml, 23.97 mmol) at 0 °C for 30 min and washed with brine (2×50 ml) and then dried over anhydrous Na_2SO_4. Concentration in vacuo gave a brown syrup, which was purified on a silica-gel column using a mixture of hexane and EtOAc (3 : 2 v/v) to give **11** as a colorless amorphous material. Yield 63% (4.34 g). $[\alpha]_D^{24}$ +5.4° (*c* 0.52, MeOH). IR 3688, 3478, 3388, 2896, 2446, 2278, 1875, 1704 cm^{-1}. ^1H NMR δ = 0.82 (d, 6H, $CH(CH_3)_2$, J = 6.4 Hz), 0.85 (d, 3H,$OCHCH_3$, J = 6.7 Hz), 0.88 (s, 9H, TPS's CH_3×3), 1.22 (t, 3H, Et's CH_3, J = 7.0 Hz), 1.41 (s, 9H, Boc), 2.20—2.30 (m, 1H, $CH(CH_3)_2$), 3.39 (s, 6H, OCH_3×2), 4.22 (q, 2H, Et's CH_2, J = 7.0 Hz), 4.33—4.40 (m, 1H, $OCHCH_3$), 4.66—4.69 (m, 1H, CHNHBoc), 5.15—5.30 (m, 1H, CHNH), 5.42 (s, 1H, $CH(OCH_3)_2$), 7.23—7.54 (m, 10H, TPS's Ph×2), 7.67 (d, 1H, Py's H, J = 8.1 Hz), 7.77 (br d, 1H, CHN*H*, J = 7.3 Hz), 8.10 (8.28 (s×2, 2H, Tz's H×2), 8.23—8.31 (m, 3H, NH, Py's H, and Tz's H). Found: C, 59.24; H, 6.10; N, 8.17%. Calcd for $C_{49}H_{60}N_6O_8S_3Si$: C, 59.73; H, 6.14; N, 8.53%.

Phenacyl 2-[2-(2-{2-[(1*S*,2*R*)-1-{2-[(*R*)-1-(*t*-Butoxycarbonylamino)-2-methylpropyl]thiazol-4-ylcarbonylamino}-2-(*t*-butyldiphenylsiloxy)propyl]thiazol-4-yl}-3-[4-(ethoxycarbonyl)thiazol-2-yl]pyridin-6-yl)thiazol-4-yl]thiazole-4-carboxylate (14). A solution of **11** (3.95 g, 3.99 mmol) in a mixture of THF and 1 M HCl (100 ml, 1 : 1) was stirred at room temperature for 12 h. Concentration in vacuo gave a residue, which was extracted with EtOAc (3×30 ml). The combined extracts were washed with brine (2×30 ml) and dried over anhydrous Na_2SO_4. Concentration in vacuo gave a yellowish amorphous substance (**12**). On the other hand, a solution of **13** (2.77 g, 5.98 mmol) in a mixture of CH_2Cl_2 and TFA (100 ml, 3 : 1) was stirred at room temperature for 1 h and then concentrated in vacuo to give a colorless syrupy residue, which was dissolved in a mixture of EtOH and H_2O (100 ml, 1 : 1) and stirred for 3 h at room temperature. EtOH and water were evaporated in vacuo to give a colorless syrup, which was dissolved in toluene (100 ml). This was mixed with a solution of **12** in toluene (100 ml) and the resulting solution was stirred for 15 min at room temperature. The reaction mixture was washed with 10% citric acid (2×50 ml), saturated aqueous solution of $NaHCO_3$ (2×50 ml), and brine (2×50 ml), and then dried over anhydrous Na_2SO_4. Concentration in vacuo gave a brownish syrup, which was again dissolved in toluene (150 ml). The solution was stirred with MnO_2 (7.64 g, 87.87 mmol) at room temperature for 12 h. The MnO_2 was filtered off and the filtrate was concentrated in vacuo to give a brown syrup. Purification on a silica-gel column using a mixture of $CHCl_3$ and acetone (15 : 1 v/v) gave crude crystals. Recrystallization from hexane–EtOAc gave **14** as pale yellow crystals. Yield 28% (1.47 g) from **11**. Mp 117—118.5 °C. $[\alpha]_D^{24}$ +11.0° (*c* 0.60, MeOH). IR 3400, 3094, 2926, 2752, 2092, 1986, 1884, 1704 cm^{-1}. ^1H NMR δ = 0.86—0.88 (m, 9H, $CH(CH_3)_2$ and O–$CHCH_3$), 0.90 (s, 9H, TPS's CH_3×3), 1.25 (t, 3H, Et's CH_3, J = 7.3 Hz), 1.41 (s, 9H, Boc), 2.20—2.30 (m, 1H, $CH(CH_3)_2$), 4.25 (q, 2H, Et's CH_2, J = 7.3 Hz), 4.35—4.45 (m, 1H, $OCHCH_3$), 4.67—4.70 (m, 1H, CHNHBoc), 5.20—5.30 (m, 1H, CHNH), 5.80 (s, 2H, Pac's CH_2), 7.20—8.04 (m, 15H, TPS's Ph×2 and Pac's Ph), 7.78 (br d, 1H, CHN*H*, J = 7.9 Hz), 8.02, 8.25, 8.30, 8.64, 8.76 (s×5, 5H, Tz's H×5), 8.37 (br d, 1H, CHN*H*, J = 8.6 Hz), 8.40 (d, 1H, Py's H, J = 8.2 Hz), 8.52 (d, 1H, Py's H, J = 8.2 Hz). Found: C, 59.28; H, 4.90; N, 8.88%. Calcd for $C_{61}H_{62}N_8O_9S_5Si$: C, 59.10; H, 5.04; N, 9.04%.

2-[2-(2-{2-[(1*S*,2*R*)-1-{2-[(*R*)-1-(*t*-Butoxycarbonylamino)-2-methylpropyl]thiazol-4-ylcarbonylamino}-2-(*t*-butyldiphenylsiloxy)propyl]thiazol-4-yl}-3-[4-(ethoxycarbonyl)thiazol-2-

yl}pyridin-6-yl)thiazol-4-yl]thiazole-4-carboxylic Acid (15). A solution of **14** (3.90 g, 3.00 mmol) and 1 M LiOH (9.01 ml, 9.01 mmol) in THF (50 ml) was stirred at 0 °C for 1 h and then acidified to pH 4 with 10% citric acid. The organic solvent of the reaction mixture was evaporated in vacuo to give an aqueous layer, which was extracted with EtOAc (3×30 ml). The combined extracts were washed with brine (2×30 ml) and then concentrated in vacuo to give a yellow syrup. Purification on a silica-gel column using EtOAc and then a mixture of $CHCl_3$ and MeOH (4 : 1 v/v) gave yellow crystals. Recrystallization from hexane–EtOAc gave **15** as pale yellow crystals. Yield 99% (3.54 g). Mp 169—170 °C. $[\alpha]_D^{24}$ −20.0° (*c* 0.03, MeOH). IR 3394, 3106, 2986, 1884, 1782, 1680, 1611 cm^{-1}. ^1H NMR (DMSO-d_6) δ = 0.84—0.86 (m, 9H, $CH(CH_3)_2$ and $OCHCH_3$), 0.87 (s, 9H, TPS's CH_3×3), 1.24 (t, 3H, Et's CH_3, *J* = 7.3 Hz), 1.41 (s, 9H, Boc), 2.20—2.30 (m, 1H, $CH(CH_3)_2$), 4.24 (q, 2H, Et's CH_2, *J* = 7.3 Hz), 4.35—4.43 (m, 1H, $OCHCH_3$), 4.60—4.70 (m, 1H, BocNHC*H*), 5.15—5.28 (m, 1H, NHC*H*), 7.17—7.53 (m, 11H, TPS's Ph×2 and Py's H), 7.77 (br d, 1H, CHN*H*, *J* = 8.2 Hz), 8.05, 8.19, 8.28, 8.29, 8.36 (s×5, 5H, Tz's H×5), 8.32 (d, 1H, Py's H, *J* = 8.2 Hz), 8.47 (br d, 1H, CHN*H*, *J* = 8.6 Hz), 8.73 (br s, 1H, COOH). Found: C, 54.92; H, 4.98; N, 9.47%. Calcd for $C_{53}H_{56}N_8O_8S_5Si{\cdot}2H_2O$: C, 55.00; H, 5.22; N, 9.68%.

(2*S*,3*R*)-2-(*t*-Butoxycarbonyl)amino-3-(*t*-butyldimethylsiloxy)butanoic Acid (17). To a solution of Boc-Thr-OBn (9.60 g, 35.90 mmol) in CH_2Cl_2 (100 ml) under cooling were added, with stirring, imidazole (6.12 g, 89.80 mmol) and TBS Cl (5.96 g, 39.50 mmol). After the mixture was stirred for 30 min and at room temperature overnight, the reaction mixture was diluted with diethyl ether (50 ml) and the resulting solution was washed with brine (3×50 ml) and dried over anhydrous Na_2SO_4. Concentration in vacuo gave a pale yellow syrup, to which was added EtOH (150 ml) and 10% Pd–C (1.1 g) at room temperature. The resulting suspension was stirred under H_2 gas stream at room temperature for 2 h and then the Pd–C was filtered off. The filtrate was concentrated in vacuo to give crude crystals, which were recrystallized from hexane–EtOAc to give **17** as colorless crystals. Yield 65% (7.72 g). Mp 108—109 °C. $[\alpha]_D^{24}$ +6.0° (*c* 0.97, MeOH). IR 3460, 3166, 2938, 1743, 1680, 1509 cm^{-1}. ^1H NMR δ = −0.03 and 0.00 (s×2, 3H×2, TBS's CH_3×2), 0.80 (s, 9H, TBS's CH_3×3), 1.04 (d, 3H, Thr's CH_3, *J* = 6.1 Hz), 1.37 (s, 9H, Boc), 3.92—3.95 (m, 1H, Thr's α-H), 4.25—4.27 (m, 1H, Thr's β-H), 5.91 (br d, 1H, CHN*H*, *J* = 9.8 Hz), 12.69 (br s, 1H, COOH). Found: C, 53.98; H, 8.93; N, 4.01%. Calcd for $C_{15}H_{31}NO_5Si$: C, 54.02; H, 9.37; N, 4.20%.

(2*S*,3*R*)-2-(*t*-Butoxycarbonyl)amino-3-(*t*-butyldimethylsiloxy)-*N*-[(*S*)-2-hydroxypropyl]butanamide (18). To a solution of (*S*)-(+)-1-aminopropan-2-ol (387 mg, 5.15 mmol) and **17** (170 mg, 5.15 mmol) in DMF (100 ml) were added, with stirring, BOP (246 mg, 5.67 mmol) and $(i\text{-Pr})_2$NEt (0.96 ml, 5.67 mmol) at 0 °C. After the mixture was stirred continuously for 30 min and at room temperature overnight, the reaction mixture was combined with H_2O (50 ml) and extracted with EtOAc (3×30 ml). The combined extracts were washed with brine (3×30 ml), 10% citric acid (2×30 ml), saturated aqueous $NaHCO_3$ solution (2×30 ml), and then dried over anhydrous Na_2SO_4. Concentration in vacuo gave a brownish syrup, which was purified on a silica-gel column using a mixture of hexane and EtOAc (1 : 1 v/v) to give **18** as a colorless syrup. Yield 90% (1.79 g). $[\alpha]_D^{24}$ +5.3° (*c* 0.5, MeOH). IR 3958, 3772, 3382, 3310, 3238, 3124, 2926, 2506, 2398, 2230, 2158, 2062, 1959, 1860, 1704, 1653, 1635, 1590 cm^{-1}. ^1H NMR δ = −0.03 and 0.00 (s×2, 6H, TBS's CH_3×2), 0.82 (s, 9H, TBS's CH_3×3), 1.01 (d, 3H, $OCHCH_3$, *J* = 6.1 Hz), 1.04 (d, 3H, Thr's CH_3, *J* = 6.1 Hz), 1.39 (s, 9H, Boc), 2.95—3.06 (m, 2H, $NHCH_2CHO$), 3.62—3.66 (m, 1H, O–$CHCH_3$), 3.91—3.94 (m, 1H, Thr's α-H), 4.10—4.14 (m, 1H, Thr's β-H), 4.65 (d, 1H, OH, *J* = 4.6 Hz), 6.07 (br d, 1H, CHN*H*, *J* = 9.5 Hz), 7.70 (br t, 1H, N*H*CH_2, *J* = 5.5 Hz). Found: C, 54.56; H, 10.07; N, 6.85%. Calcd for $C_{18}H_{38}N_2O_5Si{\cdot}1/2H_2O$: C, 54.10; H, 9.84; N, 7.01%.

(2*S*,3*R*)-2-(*t*-Butoxycarbonyl)amino-3-(*t*-butyldimethylsiloxy)-*N*-[(*S*)-2-acetoxypropyl]butanamide (19). A solution of **18** (179 mg, 4.62 mmol) in pyridine (50 ml) and acetic anhydride (50 ml) was stirred for 3 h at room temperature. Concentration in vacuo gave a brownish syrup, which was dissolved in EtOAc (50 ml). The reaction mixture was washed with brine (3×20 ml) and dried over anhydrous Na_2SO_4. Concentration in vacuo gave a pale yellow syrup, which was purified on a silica-gel column using a mixture of hexane and EtOAc (3 : 1 v/v) to give **19** as a colorless syrup. Yield 99% (1.99 g). $[\alpha]_D^{24}$ −8.2° (*c* 0.51, MeOH). IR 3820, 3664, 3574, 3328, 3076, 2926, 2506, 2452, 2254, 2128, 2032, 1908, 1671, 1587 cm^{-1}. ^1H NMR δ = −0.03 and 0.00 (s×2, 6H, TBS's CH_3×2), 0.81 (s, 9H, TBS's CH_3×3), 1.05 (d, 3H, $OCHCH_3$, *J* = 6.1 Hz), 1.11 (d, 3H, Thr's CH_3, *J* = 6.4 Hz), 1.38 (s, 9H, Boc), 1.95 (s, 3H, Ac), 3.12—3.29 (m, 2H, $NHCH_2CHO$), 3.90—3.91 (m, 1H, Thr's α-H), 4.07—4.09 (m, 1H, Thr's β-H), 4.77—4.83 (m, 1H, $OCHCH_3$), 6.06 (br d, 1H, CHN*H*, *J* = 9.6 Hz), 7.85 (br t, 1H, N*H*CH_2, *J* = 5.5 Hz). Found: C, 55.15; H, 9.81; N, 6.15%. Calcd for $C_{20}H_{40}N_2O_6Si$: C, 55.52; H, 9.32; N, 6.48%.

Ethyl 6-[4-(4-{(1*S*,2*R*)-1-[(*S*)-2-Acetoxypropylaminocarbonyl]-2-(*t*-butyldimethylsiloxy)propylaminocarbonyl}thiazol-2-yl)thiazol-2-yl]-2-(2-{2-[(1*S*,2*R*)-1-{2-[(*R*)-1-(*t*-butoxycarbonyl)amino-2-methylpropyl]thiazol-4-ylcarbonylamino}-2-(*t*-butyldiphenylsiloxy)propyl]thiazol-4-yl}pyridin-3-yl)thiazol-4-carboxylate (21). To a solution of **19** (105 mg, 2.43 mmol) in CH_2Cl_2 (30 ml) were added, with stirring, MS4A (0.5 g) and, after 10 min, TFA (15 ml) at room temperature. After the mixture was stirred for 1 h, the reaction mixture was cooled and made alkaline with Et_3N. After filtration of MS4A, the filtrate was diluted with diethyl ether (30 ml) and the resulting solution was washed with brine (2×20 ml) and dried over anhydrous Na_2SO_4. Concentration in vacuo gave a colorless syrup, which was dissolved in DMF (20 ml). After the mixture was cooled, to the resulting solution were added, with stirring, **15** (270 mg, 2.29 mmol) and $(i\text{-Pr})_2$NEt (0.58 ml, 3.43 mmol). After the mixture was stirred for 30 min under cooling and overnight at room temperature, the reaction mixture was combined with H_2O (50 ml) and extracted with EtOAc (3×30 ml). The combined extracts were washed with 10% citric acid (2×20 ml), saturated aqueous $NaHCO_3$ solution (2×20 ml), brine (3×20 ml), and then dried over anhydrous Na_2SO_4. Concentration in vacuo gave a brownish syrup, which was purified on a silica-gel column using a mixture of hexane and acetone (1 : 1 v/v) to give crude crystals. Recrystallization from hexane–EtOAc gave **21** as pale yellow crystals. Yield 87% (297 mg). Mp 112—113 °C. $[\alpha]_D^{24}$ +35.2° (*c* 0.5, MeOH). IR 3388, 2926, 2260, 1671, 1527 cm^{-1}. ^1H NMR (DMSO-d_6) δ = −0.04 and 0.00 (s×2, 6H, TBS's CH_3×2), 0.80—0.84 (m, 27H, TBS's CH_3×3, TPS's CH_3×3, $OCHCH_3$, and $CH(CH_3)_2$), 1.07—1.08 (m, 6H, Thr's CH_3 and $OCHCH_3$), 1.19 (t, 3H, Et's CH_3, *J* = 7.2 Hz), 1.35 (s, 9H, Boc), 1.90 (s, 3H, $COCH_3$), 2.15—2.25 (m, 1H, $CH(CH_3)_2$), 3.17—3.28 (m, 2H, $NHCH_2CHO$), 4.19 (q, 2H, Et's CH_2, *J* = 7.2 Hz), 4.24—4.29 (m, 1H, Thr's α-H), 4.34—4.39 (m, 2H, Thr's β-H and $OCHCH_3$), 4.61—4.64 (m, 1H, BocNHC*H*), 4.73—4.79 (m, 1H, $OCHCH_3$), 5.17—5.18 (m, 1H, NHC*H*), 7.13—7.48 (m, 11H, Ph×2, Py's H), 7.71 (br d, 1H, CHN*H*, *J* = 7.9 Hz), 7.95 (br d, 1H, CHN*H*, *J* = 8.6 Hz), 8.11

(br t, 1H, N*H*CH_2, J = 5.5 Hz), 8.17, 8.24, 8.24, 8.30, 8.34 (s×5, 5H, Tz's H×5), 8.31 (d, 1H, Py's H, J = 8.2 Hz), 8.45 (br d, 1H, CHN*H*, J = 7.9 Hz). Found: C, 56.44; H, 6.25; N, 9.41%. Calcd for $C_{68}H_{86}N_{10}O_{11}S_5Si_2$: C, 56.88; H, 6.04; N, 9.75%.

6-[4-(4-{(1*S*,2*R*)-1-[(*S*)-2-Acetoxypropylaminocarbonyl]-2-hydroxypropylaminocarbonyl}thiazol-2-yl)thiazol-2-yl]-2-(2-{2-[(1*S*,2*R*)-1-{2-[(*R*)-1-(*t*-butoxycarbonyl)amino-2-methylpropyl]thiazol-4-ylcarbonylamino}-2-(*t*-butyldiphenylsiloxy)propyl]thiazol-4-yl}pyridin-3-yl)thiazole-4-carboxylic Acid (22). A solution of **21** (297 mg, 1.99 mmol) and 70% CH_3COOH (200 ml) in THF (100 ml) was stirred for 72 h at room temperature. The reaction mixture was concentrated in vacuo to give a yellow syrup, which was dissolved in EtOAc (100 ml) and washed with saturated aqueous $NaHCO_3$ solution (2×20 ml) and brine (3×20 ml) and then dried over anhydrous Na_2SO_4. Concentration in vacuo gave a yellow syrup, which was purified on a silica-gel column using a mixture of hexane and acetone (1 : 2 v/v) to give crude crystals. Recrystallization from hexane–EtOAc gave **22** as pale yellow crystals. Yield 64% (176 mg). Mp 116.0—117.5 °C. $[\alpha]_D^{24}$ +35.9° (*c* 0.69, MeOH). IR 3718, 3406, 3094, 2938, 2242, 1983, 1788, 1653 cm^{-1}. ^1H NMR (DMSO-d_6) δ = 0.86—0.90 (m, 18H, TPS's CH_3×3, OCHCH_3, and CH(CH_3)$_2$), 1.11 (d, 3H, Thr's CH_3, J = 6.4 Hz), 1.13 (d, 3H, OCHCH_3, J = 6.4 Hz), 1.25 (t, 3H, Et's CH_3, J = 7.0 Hz), 1.41 (s, 9H, Boc), 1.97 (s, 3H, $COCH_3$), 2.20—2.30 (m, 1H, C*H*(CH_3)$_2$), 3.19—3.36 (m, 2H, NHCH_2CHO), 4.08—4.13 (m, 1H, Thr's β-H), 4.25 (q, 2H, Et's CH_2, J = 7.0 Hz), 4.37—4.40 (m, 2H, Thr's α-H and OC*H*CH_3), 4.66—4.69 (m, 1H, BocNHC*H*), 4.81—4.86 (m, 1H, O–C*H*CH_3), 5.10 (d, 1H, OH, J = 5.2 Hz), 5.22—5.24 (m, 1H, NHC*H*), 7.20—7.55 (m, 10H, TPS's Ph×2), 7.79 (br d, 1H, CHN*H*, J = 8.6 Hz), 8.06 (br d, 1H, CHN*H*, J = 8.6 Hz), 8.14 (br t, 1H, N*H*CH_2, J = 5.8 Hz), 8.25, 8.30, 8.30, 8.42, 8.61 (s×5, 5H, Tz's H×5), 8.31—8.40 (m, 1H, Py's H), 8.39 (br d, 1H, CHN*H*, J = 8.5 Hz), 8.51 (d, 1H, Py's H, J = 8.2 Hz). Found: C, 56.76; H, 5.63; N, 10.29%. Calcd for $C_{62}H_{72}N_{10}O_{11}S_5Si$: C, 56.34; H, 5.49; N, 10.60%.

Ethyl 6-{4-[4-({(*Z*)-1-[(*S*)-2-Acetoxypropylaminocarbonyl]-1-propenyl}aminocarbonyl)thiazol-2-yl]thiazol-2-yl}-2-(2-{2-[(1*S*,2*R*)-1-{2-[(*R*)-1-(*t*-butoxycarbonyl)amino-2-methylpropyl]thiazol-4-ylcarbonylamino}-2-(*t*-butyldiphenylsiloxy]propylthiazol-4-yl}pyridin-3-yl)thiazole-4-carboxylate (23). To a solution of **22** (159 mg, 4.63 mmol) in DMSO (50 ml) were added, with stirring, Et_3N (0.35 ml, 2.53 mmol) and MsCl (0.098 ml, 1.26 mmol) at 0 °C and, after stirring for 30 min, DBU (0.19 ml, 1.26 mmol). After the mixture was stirred overnight at room temperature, the reaction mixture was combined with EtOAc (100 ml) and washed with 10% citric acid (2×100 ml), saturated aqueous $NaHCO_3$ solution (2×100 ml), and brine (2×100 ml) and then dried over anhydrous Na_2SO_4. Concentration in vacuo gave a yellow syrup, which was purified on a silica-gel column using a mixture of hexane and EtOAc (2 : 3 v/v) to give **23** as pale yellow crystals. Yield 97% (152 mg). Mp 117.0—118.5 °C. $[\alpha]_D^{24}$ +12.3° (*c* 0.69, MeOH). IR 3406, 2962, 2230, 1674 cm^{-1}. ^1H NMR δ = 0.86—0.88 (m, 9H, OCHCH_3 and CH(CH_3)$_2$), 0.90 (s, 9H, TPS's CH_3×3), 1.13 (d, 3H, OCHCH_3, J = 6.4 Hz), 1.16 (t, 3H, Et's CH_3, J = 7.0 Hz), 1.41 (s, 9H, Boc), 1.71 (d, 3H, CH_3CH=, J = 7.0 Hz), 1.97 (s, 3H, $COCH_3$), 2.20—2.30 (m, 1H, C*H*(CH_3)$_2$), 3.19—3.36 (m, 2H, NH CH_2CHO), 4.24 (q, 2H, Et's CH_2, J = 7.0 Hz), 4.39—4.41 (m, 1H, OC*H*CH_3), 4.67—4.70 (m, 1H, BocNHC*H*), 4.85—4.90 (m, 1H, OC*H*CH_3), 5.23—5.26 (m, 1H, NHC*H*), 6.45 (q, 1H, CH_3C*H*=, J = 7.0 Hz), 7.20—7.56 (m, 10H, TPS's Ph×2), 7.79 (br d, 1H, CHN*H*, J = 7.9 Hz), 8.10 (br t, 1H, N*H*CH_2, J = 5.8 Hz), 8.24, 8.29, 8.30, 8.45, 8.61 (s×5, 5H, Tz's H×5), 8.36—8.38 (m, 1H, Py's H), 8.38 (br d, 1H, CHN*H*, J = 7.9 Hz), 8.51 (d, 1H, Py's H, J = 8.2 Hz), 9.48 (br s, 1H, NH). Found: C, 56.84; H, 5.51; N, 10.62%. Calcd for $C_{62}H_{70}N_{10}O_{10}S_5Si$: C, 57.12; H, 5.42; N, 10.75%.

2-(2-{2-[(1*S*,2*R*)-1-{2-[(*R*)-1-(*t*-Butoxycarbonyl)amino-2-methylpropyl]thiazol-4-ylcarbonylamino}-2-(*t*-butyldiphenylsiloxy)propyl]thiazol-4-yl}-6-({4-[4-{(*Z*)-1-[(*S*)-2-hydroxypropylaminocarbonyl]-1-propenyl}aminocarbonyl)thiazol-2-yl]thiazol-2-yl}pyridin-3-yl)thiazole-4-carboxylic Acid (24). To a solution of **23** (198 mg, 1.46 mmol) in MeOH–H_2O (50 ml, 3 : 2 v/v) was added, with stirring, 1M LiOH (2.25 ml, 2.25 mmol) at 0 °C. After the mixture was stirred at room temperature overnight, the reaction mixture was acidified to pH 3—4 with citric acid and then concentrated in vacuo to give a residual yellow syrup. The syrup was dissolved in EtOAc (50 ml) and washed with brine (2×20 ml). Concentration in vacuo gave crude crystals, which were recrystallized from hexane–EtOAc to give **24** as pale yellow crystals. Yield 72% (136 mg). Mp 204—205 °C. $[\alpha]_D^{24}$ −0.6° (*c* 0.33, MeOH). IR 3376, 2956, 1995, 1887, 1656 cm^{-1}. ^1H NMR δ = 0.85—0.89 (m, 9H, OCHCH_3 and CH(CH_3)$_2$), 0.89 (s, 9H, TPS's CH_3×3), 1.06 (d, 3H, OCHCH_3, J = 6.4 Hz), 1.40 (s, 9H, Boc), 1.68 (d, 3H, CH_3CH=, J = 7.0 Hz), 2.20—2.30 (m, 1H, C*H*(CH_3)$_2$), 3.02—3.12 (m, 2H, NHCH_2CHO), 3.69—3.72 (m, 1H, OC*H*CH_3), 4.30—4.40 (m, 1H, OC*H*CH_3), 4.60—4.75 (m, 2H, OH and BocNHC*H*), 5.10—5.25 (m, 1H, NHC*H*), 6.50 (q, 1H, CH_3C*H*=, J = 7.0 Hz), 7.15—7.55 (m, 11H, TPS's Ph×2 and Tz's H), 7.80 (br d, 1H, CHN*H*, J = 7.9 Hz), 7.90—8.00 (m, 1H, Py's H), 8.33, 8.44, 8.58, 9.11 (s×4, 4H, Tz's H×4), 8.20—8.70 (m, 1H, COOH), 8.30—8.40 (m, 1H, Py's H), 8.52 (br d, 1H, CHN*H*, J = 8.2 Hz), 9.50 (br s, 1H, NH). Found: C, 53.17; H, 5.14; N, 10.19%. Calcd for $C_{58}H_{63}N_{10}O_9S_5Si\cdot 4H_2O$: C, 53.39; H, 4.87; N, 10.73%.

Ethyl 2-{(*Z*)-1-[(2*S*,3*R*)-{2-[2-(2-{2-[(1*S*,2*R*)-1-{2-[(*R*)-1-(*t*-Butoxycarbonylamino)-2-methylpropyl]thiazol-4-ylcarbonylamino}-2-(*t*-butyldiphenylsiloxy)propyl]thiazol-4-yl}-6-{4-[4-((*Z*)-{1-[(*S*)-2-hydroxypropylaminocarbonyl]-1-propenyl}aminocarbonyl)thiazol-2-yl]thiazol-2-yl}pyridin-3-yl)thiazol-4-ylcarbonylamino]-3-(*t*-butyldiphenylsiloxy)}butanoylamino]-1-propenyl}thiazole-4-carboxylate (26). To a solution of **25** (106 mg, 1.58 mmol) in CH_2Cl_2 (30 ml) were added MS4A (0.5 g) and TFA (15 ml) for 10 min under cooling. After the mixture was stirred for 1 h under cooling, the resulting solution was made alkaline with Et_3N. 4AMS was filtered off and the filtrate was combined with diethyl ether (30 ml) and then washed with brine (2×20 ml) and finally dried over anhydrous Na_2SO_4. Concentration in vacuo gave a colorless syrup, which was dissolved in DMF (20 ml) and cooled. To the resulting solution were added, with stirring, **24** (136 mg, 1.05 mmol), BOP (502 mg, 1.16 mmol) and (i-Pr)$_2$NEt (0.20 ml, 1.16 mmol) for 30 min under cooling and overnight at room temperature. The reaction mixture was poured into water (50 ml) and then extracted with EtOAc (3×20 ml). The combined extracts were washed with 10% citric acid (2×10 ml), saturated aqueous $NaHCO_3$ solution (2×10 ml), and brine (3×10 ml) and then dried over anhydrous Na_2SO_4. Concentration in vacuo gave a brownish syrup, which was purified on a silica-gel column using a mixture of $CHCl_3$ and acetone (4 : 1 v/v) to give crude crystals. Recrystallization from hexane–EtOAc gave **26** as colorless crystals. Yield 91% (196 mg). Mp 132.0—133.5 °C. $[\alpha]_D^{24}$ +11.0° (*c* 0.49, MeOH). IR 3370, 3064, 2932, 2248, 1962, 1659 cm^{-1}. ^1H NMR δ = 0.85—0.91 (m, 12H, TPS's CH_3×3 and O–CHCH_3), 0.87 (d, 6H, CH(CH_3)$_2$, J = 6.4 Hz), 0.97 (s, 9H, TPS's CH_3×3), 1.01—1.03 (m, 6H, Thr's CH_3 and OCHCH_3), 1.26 (t, 3H, Et's CH_3, J = 7.3 Hz), 1.41 (s, 9H, Boc), 1.66 (d, 3H, CH_3CH=, J = 7.0 Hz),

1.69 (d, 3H, Pp's CH_3, J = 7.0 Hz), 2.20—2.30 (m, 1H, C*H*$(CH_3)_2$), 3.03—3.13 (m, 2H, NHCH_2CHO), 3.69—3.73 (m, 1H, OC*H*CH_3), 4.27 (q, 2H, Et's CH_2, J = 7.3 Hz), 4.53—4.57 (m, 2H, OC*H*CH_3 and Thr's β-H), 4.62 (d, 1H, OH, J = 4.4 Hz), 4.65—4.75 (m, 2H, Thr's α-H and BocNHC*H*), 5.27—5.29 (m, 1H, NHC*H*), 6.50 (q, 1H, CH_3C*H*=, J = 7.0 Hz), 6.59 (q, 1H, CH_3C*H*=, J = 7.0 Hz), 7.16—7.66 (m, 20H, TPS's Ph×4), 7.77 (br d, 1H, CHN*H*, J = 7.6 Hz), 7.89 (br t, 1H, N*H*CH_2, J = 5.5 Hz), 8.21, 8.26, 8.33, 8.36, 8.45, 8.60 (s×6, 6H, Tz's H×6), 8.25—8.30 (m, 2H, CHN*H* and Py's H), 8.50 (br d, 1H, CHN*H*, J = 8.2 Hz), 8.54 (d, 1H, Py's H, J = 8.2 Hz), 9.47 (br s, 1H, NH), 9.89 (br s, 1H, NH). Found: C, 58.52; H, 5.65; N, 10.06%. Calcd for $C_{88}H_{99}N_{13}O_{12}S_6Si_2 \cdot H_2O$: C, 58.80; H, 5.66; N, 10.13%.

Micrococcin P_1 (1). To a solution of **26** (697 mg, 0.392 mmol) in $CHCl_3$ (20 ml) was added TFA (20 ml) and the resulting solution was stirred for 3 h at room temperature and then concentrated in vacuo to give a pale yellow syrup. The syrup was dissolved in MeOH (50 ml) and 1M LiOH (39.20 ml, 39.2 mmol) was added to the MeOH solution at 0 °C. After this was stirred for 30 min at 0 °C and overnight at room temperature, the reaction mixture was adjusted to pH 7 with Dowex 50H$^+$ and the ionic exchange resin was filtered off. The filtrate was concentrated in vacuo to give crude yellowish crystals, which were dissolved in DMF (100 ml). To the resulting cooled solution were added, with stirring, BOP (187 mg, 0.431 mmol) and (i-Pr)$_2$NEt (0.073 ml, 0.431 mmol), and then the resulting solution was stirred continuously for 1 h under cooling and overnight at room temperature. Concentration in vacuo gave a yellowish syrup, which was dissolved in EtOAc (50 ml) and washed with brine (2×10 ml) and then dried over anhydrous Na_2SO_4. Concentration again gave a brownish syrup, which was purified on a silica-gel column using a mixture of $CHCl_3$ and MeOH (20 : 1 v/v) to give colorless crystals. TBAF (310 mg, 0.118 mmol) was added, with stirring, to a solution of the obtained crystals in THF (20 ml) at room temperature. After the mixture was stirred for 3 h, the reaction mixture was concentrated in vacuo to give crude brownish crystals, which were purified on a TLC using a mixture of $CHCl_3$ and MeOH (15 : 1 v/v) to give **1** as colorless crystals. Yield 13% (52 mg). Mp 210—245 °C. $[\alpha]_D^{24}$ +40.0° (c 0.50, 90% EtOH). λ_{max} (nm) 324.6. IR 3418, 1653, 1527 cm^{-1}. ^{1}H NMR ($CDCl_3$–CD_3OD) δ = 0.78 (d, 3H, OCHC*H*$_3$, J = 6.5 Hz), 1.15 (d, 3H, OCHC*H*$_3$, J = 6.5 Hz), 1.19 (d, 3H, CH(C*H*$_3$)$_2$, J = 7.0 Hz), 1.31 (d, 3H, Thr's CH_3, J = 7.0 Hz), 1.34 (d, 3H, Thr's CH_3, J = 6.5 Hz), 1.83 and 1.86 (d, 3H, C*H*$_3$CH=, J = 7.0 Hz), 2.82—2.92 (m, 1H, C*H*$(CH_3)_2$), 3.14—3.37 (m, 2H, NHCH_2CHO), 3.88—3.91 (m, 1H, OC*H*CH_3), 4.09—4.11 (m, 1H, OC*H*CH_3), 4.52—4.53 (m, 1H, C*H*NH), 4.54—4.56 (m, 1H, Thr's β-H), 4.71—4.77 (m, 1H, Thr's α-H), 5.18 (d, 1H, C*H*NH, J = 9.0 Hz), 6.57 (q, 1H, CH_3C*H*=, J = 7.0 Hz), 6.71 (q, 1H, CH_3C*H*=, J = 7.0 Hz), 7.78, 7.98, 8.08, 8.25, 8.29, 8.41 (each s, 6H, Tz's H×6), 8.17 and 8.34 (d, 1H, Py's H, J = 8.5 Hz). ^{1}H NMR (DMSO-d_6) δ = 0.91 (d, 3H, OCHC*H*$_3$, J = 6.1 Hz), 0.96 (d, 3H, CHC*H*$_3$, J = 6.7 Hz), 1.01 (d, 3H, OCHC*H*$_3$, J = 6.1 Hz), 1.12 (d, 3H, CHC*H*$_3$, J = 6.7 Hz), 1.19 (d, 3H, Thr's CH_3, J = 7.0 Hz), 1.69 (d, 3H, C*H*$_3$CH=, J = 7.0 Hz), 1.79 (d, 3H, C*H*$_3$CH=, J = 7.0 Hz), 2.55—2.70 (m, 1H, C*H*$(CH_3)_2$), 3.02—3.12 (m, 2H, NHCH_2CHO), 3.68—3.72 (m, 1H, OC*H*CH_3), 3.99—4.03 (m, 1H, OC*H*CH_3), 4.07 (br s, 1H, OH), 4.23—4.25 (m, 1H, Thr's β-H), 4.45—4.50 (m, 1H, Thr's α-H), 4.63 (br s, 1H, OH), 5.01—5.03 (m, 1H, C*H*NH), 5.12—5.16 (m, 1H, C*H*NH), 6.37 (br s, 1H, CH_3C*H*=), 6.49 (q, 1H, CH_3C*H*=, J = 7.0 Hz), 7.84 (br d, 1H, CHN*H*, J = 7.3 Hz), 7.89 (br t, 1H, N*H*CH_2, J = 5.5 Hz), 8.10—8.20 (m, 1H, Thr's NH), 8.47 (br d, 1H, NH, J = 9.5 Hz), 8.33 (d, 1H, Py's H, J = 8.2 Hz), 8.47 (d, 1H, Py's H, J = 8.2 Hz), 7.98, 8.15, 8.20, 8.32, 8.45, 8.60 (each s, 6H, Tz's H×6), 9.32 (br s, 1H, NH), 9.49 (br s, 1H, NH). ^{13}C NMR ($CDCl_3$–CD_3OD) δ = 12.1, 14.0, 19.2, 20.6, 20.7, 20.7, 21.2, 33.1, 47.8, 55.1, 59.2, 60.7, 67.0, 67.3, 69.2, 119.1, 121.6, 122.5, 124.4, 125.0, 126.1, 126.6, 129.4, 130.3, 130.5, 131.6, 131.6, 140.9, 148.6, 149.7, 150.1, 150.6, 150.8, 151.6, 152.1, 153.2, 161.5, 162.1, 162.8, 163.3, 163.6, 166.8, 167.2, 167.5, 169.3, 169.7, 172.3, 172.7. ^{13}C NMR (DMSO-d_6) δ = 13.5, 13.6, 19.5, 19.6, 19.8, 20.6, 21.0, 32.0, 46.9, 56.2, 57.0, 59.0, 65.1, 66.5, 67.6, 118.5, 121.7, 122.7, 123.9, 124.8, 125.4, 126.3, 127.7, 127.8, 128.7, 130.1, 130.6, 140.2, 148.2, 148.6, 148.7, 149.4, 149.9, 150.4, 150.7, 151.1, 159.0, 160.1, 160.5, 160.5, 161.3, 164.1, 164.3, 166.2, 168.4, 168.9, 169.2, 171.0.

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