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Asymmetric Total Synthesis of (−)-Saframycin A from L-Tyrosine

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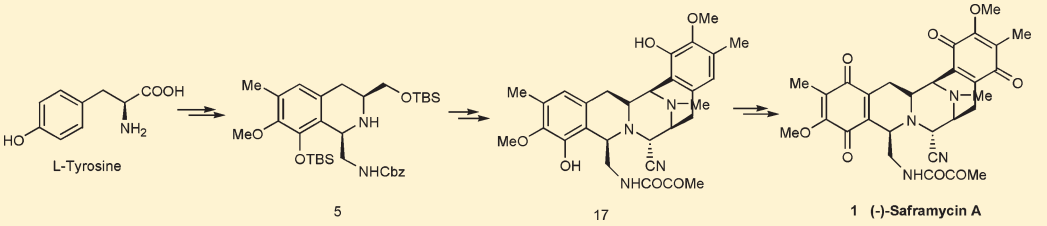
Supporting Information

ABSTRACT:



The asymmetric total synthesis of (−)-saframycin A, a natural antitumor product of the tetrahydroisoquinoline antitumor antibiotics family, has been accomplished by employing L-tyrosine as the starting chiral building block in 24 steps for the longest linear sequence in an overall yield of 9.7%. The key steps in the synthesis involve stereoselective intermolecular and intramolecular Pictet−Spengler reactions, which induced the correct stereochemistry at C-1 and C-11, respectively. The selective protection−deprotection protocol of an amino group in the two-step transformation from intermediate **10** to **12** and a hydroxyl group in the first two steps resulted in both high selectivity and efficiency of the synthetic route.

INTRODUCTION

(−)-Saframycin A is a member of the tetrahydroisoquinoline antitumor antibiotics that were isolated from *Streptomyces lavendulae* 314 in 1977 by Arai et al.[1−3] Members of this family of alkaloids have shown potent antitumor activities, such as Et-743, which has been launched in Europe for treating soft tissue sarcoma,[4] and its analogue Zalypsis, which has been under phase II clinical trials for treating Ewing's sarcoma[5,6] (Figure 1). As a typical member of this family of natural products, (−)-saframycin A has a characteristic pentacyclic skeleton and remarkable antitumor activity. So far, one racemic and two asymmetric total syntheses have been reported.[7−11]

Our group has established a novel biomimetic strategy for the stereocontrolled synthesis of the tetrahydroisoquinoline alkaloids employing L-dopa or L-tyrosine as the starting chiral building block. A series of simplified analogues have been synthesized from L-dopa,[12,13] and (−)-renieramycin G and its derivatives have been synthesized from L-tyrosine in our lab.[14−16] As a continuation of the total synthesis research on the tetrahydroisoquinoline antitumor antibiotics, we report herein a biomimetic total synthesis of (−)-saframycin A from L-tyrosine.

RESULTS AND DISCUSSION

The retrosynthetic analysis of (−)-saframycin A is shown in Scheme 1. We envisioned that (−)-saframycin A (**1**) could be obtained by the oxidation of the phenolic hydroxyl groups of compound **17**. Next, the pentacyclic skeleton could be constructed through the stereospecific intramolecular Pictet−Spengler reaction of compound **10**. Compound **10** could be synthesized through the coupling of *N*-Cbz-protected 1,2,3,4-tetrahydroisoquinoline moiety **5** and *N*-Boc protected amino acid moiety **6**, both of which could be prepared through several steps from the same starting material L-tyrosine.

Following the strategy of our previous studies on bistetrahydroisoquinoline alkaloids, we obtained amino acid moiety **6** from commercially available L-tyrosine methyl ester in 12 steps in an overall yield of 36%.[14] On the other hand, the amino alcohol intermediate **2** and *N*-Cbz-protected amino acetaldehyde **3** were coupled in acetic acid through the stereospecific intermolecular Pictet−Spengler reaction to afford compound **4** in 86% yield (Scheme 2).[15] In this reaction, the use of bromine to protect the position *para* to the hydroxyl group was avoided to improve the synthetic efficiency. The regioselectivity and stereoselectivity for the conversion of **2** to **4** were found to be affected by the temperature and the addition speed of aldehyde **3**. Keeping a slow addition speed and the temperature at 0 °C, the 1,3-*cis*-stereoisomer could be obtained dominantly through the highly regioselective and diastereoselective *ortho* cyclization on the benzene ring. Strong NOE correlations between Me- and H-5 indicated that a correct cyclization occurred at the *ortho* site of the phenolic hydroxyl group. Additionally, obvious NOE correlations between H-1 and H-3 confirmed a *cis* relationship between these two protons (Figure 2). Silylation of compound

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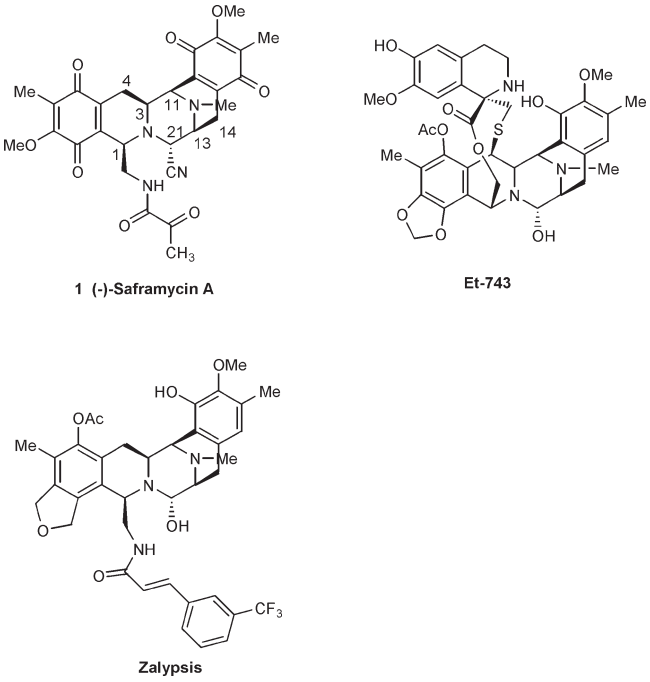
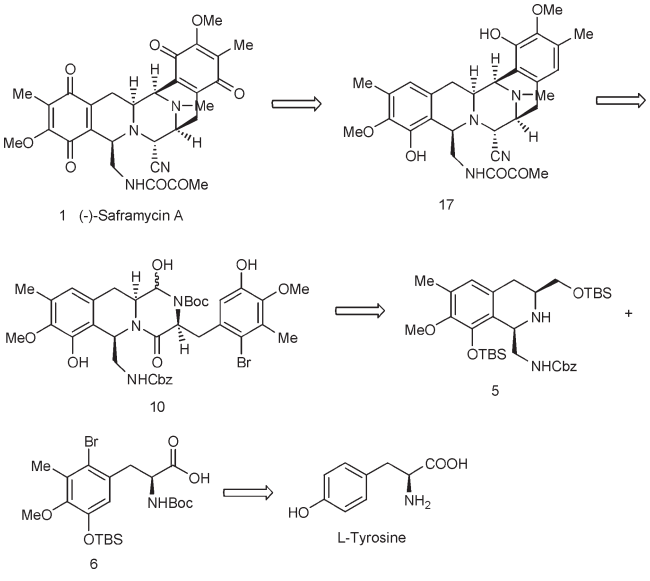


Figure 1. Structures of tetrahydroisoquinoline alkaloids.

Scheme 1. Retrosynthetic Analysis of (−)-Saframycin A



4 with TBSCl afforded *O*-TBS-protected 1,2,3,4-tetrahydroisoquinoline fragment **5**.[17]

1,2,3,4-Tetrahydroisoquinoline **5** and amino acid **6** were coupled through the catalysis of BOPCl to give the amide intermediate **7**.[18] Then the TBS-ether derivative **7** was selectively deprotected with HCOOH to afford the primary alcohol compound **8**.[19] Oxidation of compound **8** with Dess−Martin periodinane afforded the corresponding hemiaminal **9**.[20] Complete removal of the two phenolic TBS groups of compound **9** with TBAF afforded compound **10**. Having obtained the deprotected hemiaminal **10**, we investigated its stereoselective conversion to the pentacyclic skeleton intermediate **11**.[14,21] On the basis of the already established intramolecular Pictet−Spengler reaction conditions by our group, super acid TfOH (trifluoromethanesulfonic acid) was chosen to realize the Pictet−Spengler cyclization reaction. However, several attempts using various proportions of TfOH in CH_2Cl_2 were not successful, and only N-deprotected **10** was obtained. Finally, pure TfOH was used to treat compound **10** at room temperature for two hours to successfully give the expected pentacyclic intermediate **11** in 83% yield. Noticeably, the *N*-Boc and *N*-Cbz protecting groups were removed simultaneously in this reaction. Next, the primary amine group of compound **12**. Reductive methylation of compound **12** with HCHO afforded compound **13** (Scheme 3).

With intermediate **13** in hand, we completed the final stage of the synthesis of (−)-saframycin A in five steps. First, removal of the bromine atom by catalytic hydrogenolysis afforded compound **14**. Then the lactam ring of compound **14** was reduced with $LiAlH_4$ at low temperature in THF for two hours to afford the corresponding hemiaminal intermediate, which was then cyanided with KCN in acidified THF/H_2O to give amino nitrile **15**.[22] The *N*-Boc protecting group of compound **15** was removed in high yield via treatment with TFA. Then deprotected compound **16** was coupled with pyruvic acid to afford pyruvamide **17**.[23] Oxidation of compound **17** with air in the presence of catalyst salcomine afforded (−)-saframycin A as the final target product (Scheme 4).[24] The ^{1}H NMR, ^{13}C NMR, and HRMS spectra and the specific optical rotation of the synthetic product **1** were consistent with those reported for natural saframycin A.[1,2,11]

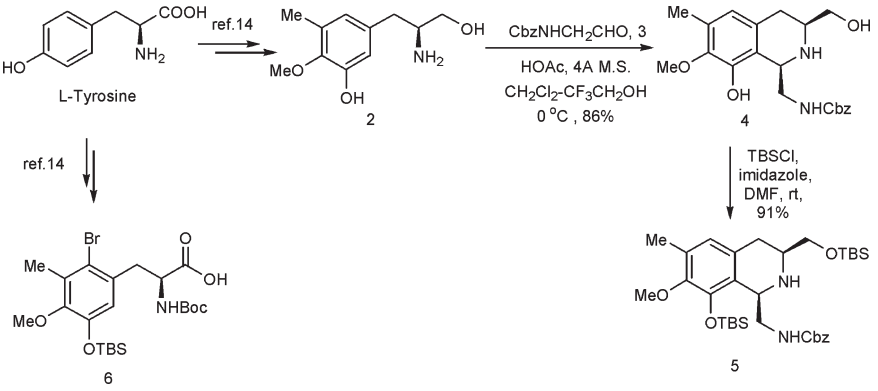
CONCLUSIONS

(−)-Saframycin A has been successfully synthesized as a single enantiomer in 24 steps for the longest linear sequence from L-tyrosine methyl ester in an overall yield of 9.7%. In the key step of the synthesis of 1,2,3,4-tetrahydroisoquinoline **5**, we avoided introducing the bromine atom at the *para* site of the phenolic hydroxyl group and realized the highly regioselective and stereoselective cyclization through the careful control of the temperature and the addition speed of the aldehyde. The key pentacyclic intermediate 11 was constructed through the intramolecular Pictet−Spengler reaction with pure TfOH as both the solvent and catalyst. Following a selective protection−deprotection protocol, we finally obtained (−)-saframycin A which is identical with the natural product. Among the five chiral centers, C-3 and C-13 came from L-tyrosine; C-1 and C-11 were induced through the intermolecular and intramolecular Pictet−Spengler reaction; and C-21 was induced in the substitution reaction. In the future studies, the present route with L-tyrosine as the starting chiral building block will be employed to synthesize the derivatives of (−)-saframycin A and other related tetrahydroisoquinoline antitumor antibiotics.

EXPERIMENTAL SECTION

Benzyl((1*R*,3*S*)-8-hydroxy-3-(hydroxymethyl)-7-methoxy-6-methyl-1,2,3,4-tetrahydroisoquinolin-1-yl)methylcarbamate (4). To a stirred solution of compound **2** (6.90 g, 32.70 mmol), acetic acid (4.98 mL, 81.75 mmol), and 4 Å molecular sieves (5.00 g) in CH_2Cl_2−CF_3CH_2OH (7:1, v/v, 180 mL) was added aldehyde **3** (0.6 M in CH_2Cl_2, 60 mL) dropwise at 0 °C under argon, and then the mixture was stirred for 10 h at this temperature. The reaction mixture was filtered and concentrated under reduced pressure to give the crude product. Purification by column chromatography (CH_2Cl_2/MeOH = 100:3) afforded the tetrahydroisoquinoline product **4** (10.8 g, 86%) as a white solid.

Scheme 2. Synthesis of Compounds 5 and 6



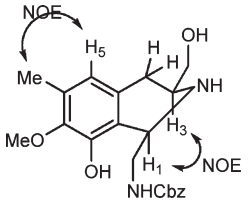
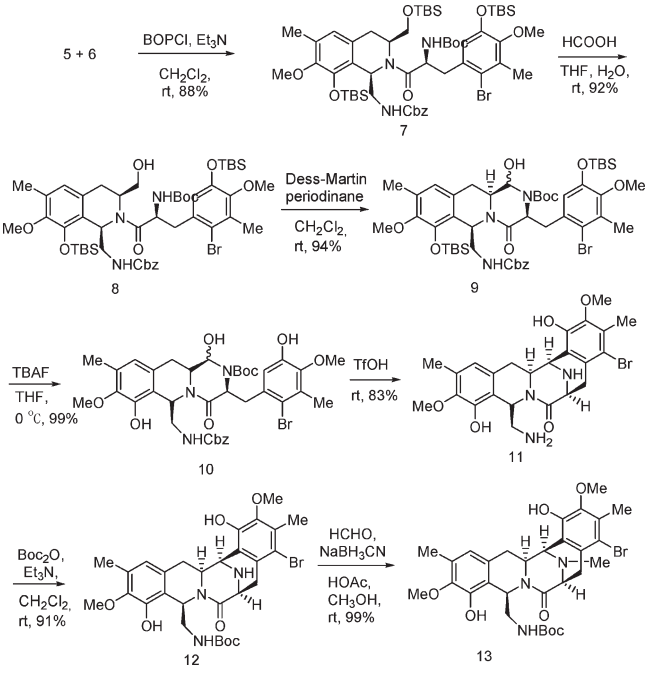


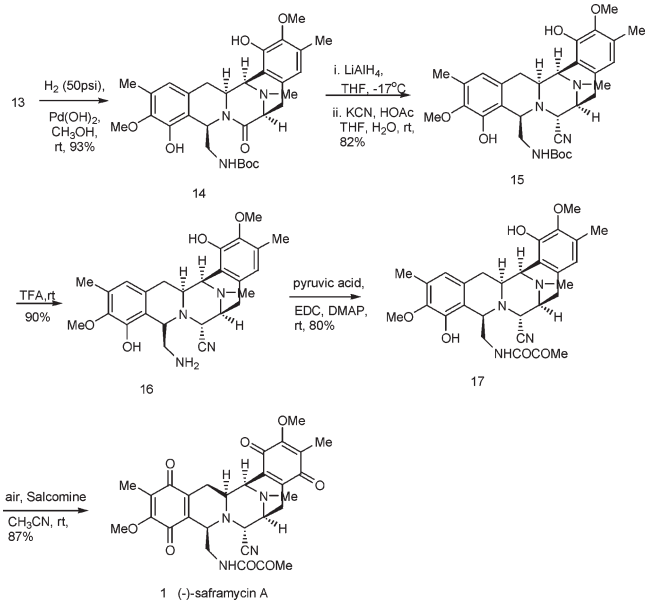
Figure 2. NOE correlations of compound **4**.

Scheme 3. Synthesis of Compound 13



Mp = 157−159 °C; $[\alpha]_D^{20}$ = −137 (*c* 0.10, CH_2Cl_2); ^{1}H NMR (600 MHz, DMSO-d_6) δ 8.69 (s, 1H), 7.36−7.29 (m, 5H), 6.91 (t, *J* = 5.4 Hz, 1H), 6.37 (s, 1H), 4.99 (d, *J* = 12.6 Hz, 2H), 4.61 (t, *J* = 5.4 Hz, 1H), 4.20 (d, *J* = 6.0 Hz, 1H), 3.74−3.71 (m, 1H), 3.59 (s, 3H), 3.42−3.39 (m, 1H), 3.34−3.30 (m, 1H), 3.06−3.01 (m, 1H), 2.69−2.68 (m, 1H), 2.42 (dd, *J* = 14.4, 1.8 Hz, 1H), 2.25 (dd, *J* = 14.4, 10.8 Hz, 1H), 2.14 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 156.3, 146.6, 143.9, 137.3, 132.5, 128.3 (3C), 128.0, 127.6 (2C), 121.7, 120.8, 65.4, 65.0, 59.9, 54.1, 52.2, 45.9, 32.8, 15.4; HRMS calcd for $C_{21}H_{27}N_2O_5$ $[M + H]^+$ 387.1919, found 387.1920.

Scheme 4. Synthesis of (−)-Safranycin A



Benzyl((1*R*,3*S*)-8-(*tert*-butyldimethylsilyloxy)-3-((*tert*-butyldimethylsilyloxy)methyl)-7-methoxy-6-methyl-1,2,3,4-tetrahydroisoquinolin-1-yl)methylcarbamate (5). To a solution of compound **4** (5.00 g, 12.95 mmol) in DMF (15 mL) was added imidazole (7.78 g, 113.96 mmol) and TBSCl (8.60 g, 56.98 mmol), and then the mixture was stirred at room temperature under argon for 48 h. After dilution with EtOAc (150 mL), the mixture was washed with water (70 mL × 3) and brine (70 mL), dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (PET/EtOAc = 10:1) afforded the TBS-protected product **5** (7.25 g, 91%) as a yellow oil. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.36−7.28 (m, 5H), 6.54 (s, 1H), 5.08−5.00 (m, 2H), 4.43 (s, 1H), 3.68 (d, *J* = 9.9 Hz, 1H), 3.64 (d, *J* = 9.9 Hz, 1H), 3.59 (s, 3H), 3.60−3.54 (m, 2H), 2.92−2.82 (m, 1H), 2.46−2.41 (m, 2H), 2.21 (s, 3H), 0.97 (s, 9H), 0.91 (s, 9H), 0.26 (s, 3H), 0.09 (s, 9H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 156.6, 147.8, 145.8, 136.8, 132.7, 129.9, 128.4 (2C), 127.9 (2C), 125.6, 124.4, 123.8, 67.3, 66.8, 66.3, 60.0, 54.0, 52.8, 45.4, 32.8, 26.1 (2C), 25.9 (2C), 25.6, 18.6, 18.4, 15.8, −3.6, −3.7, −4.3, −5.3; HRMS calcd for $C_{33}H_{55}N_2O_5Si_2$ $[M + H]^+$ 615.3649, found 615.3645.

Compound 7. To an ice-cooled solution of compound **5** (8.90 g, 14.50 mmol) in CH_2Cl_2 (200 mL) were added Et_3N (5.20 mL, 37.70

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mmol), compound **6** (9.74 g, 18.85 mmol), and BOPCl (4.80 g, 18.85 mmol) in portions, and then the mixture was stirred at room temperature for 72 h. The reaction was quenched with 2 N aq. HCl, and the organic phase was separated and washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (PET/EtOAc = 20:1) afforded the amide derivative **7** (14.23 g, 88%) as a white solid. Mp = 62−64 °C; $[\alpha]_D^{20}$ = +10 (*c* 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.31−7.29 (m, 5H), 6.58 (s, 1H), 6.55 (s, 1H), 5.99 (m, 1H), 5.89 (br s, 1H), 5.06 (s, 2H), 4.37 (m, 1H), 3.86−3.74 (m, 2H), 3.73 (m, 1H), 3.71 (m, 1H), 3.63 (s, 3H), 3.62 (s, 3H), 3.24−3.14 (m, 1H), 3.00−2.90 (m, 1H), 2.90−2.80 (m, 1H), 2.78−2.70 (m, 1H), 2.70−2.62 (m, 1H), 2.35 (s, 1H), 2.27 (s, 3H), 2.22 (s, 3H), 1.29 (s, 9H), 1.02 (s, 9H), 0.98 (s, 9H), 0.84 (s, 9H), 0.28 (s, 3H), 0.17 (s, 3H), 0.16 (s, 3H), 0.14 (s, 3H), 0.05 (s, 3H), 0.03 (s, 3H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 174.0, 156.2, 154.6, 149.1, 148.1, 147.5, 145.1, 137.1, 132.8, 131.6, 131.1, 128.3 (2C), 127.8, 127.7 (2C), 127.6, 124.2, 123.3, 121.9, 119.2, 79.1, 68.0, 66.1, 65.4, 60.0, 52.9, 52.3, 50.6, 48.8, 45.6, 41.0, 28.8, 28.2 (3C), 26.1 (3C), 25.9 (3C), 25.6 (3C), 18.6, 18.2, 18.1, 16.9, 15.8, −3.8, −4.3, −4.6, −5.5, −5.6; HRMS calcd for $C_{55}H_{89}BrN_3O_{10}Si_3$ $[M + H]^+$ 1114.5033, found 1114.4994.

Compound 8. A mixture of compound **7** (8.60 g, 7.73 mmol) in THF−HCOOH−H_2O (6:3:1, 130 mL) was stirred at room temperature for 10 h, and then the mixture was concentrated under reduced pressure. The residue was dissolved in EtOAc, washed with saturated aq. $NaHCO_3$ and brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (PET/EtOAc = 8:1) afforded selectively deprotected product **8** (7.11 g, 92%) as a white solid. Mp = 78−80 °C; $[\alpha]_D^{20}$ = +21 (*c* 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, DMSO-d_6) δ 7.33−7.14 (m, 5H), 6.87 (s, 1H), 6.63 (s, 1H), 6.30 (s, 1H), 5.92 (s, 1H), 4.99 (d, *J* = 12.6 Hz, 1H), 4.92 (d, *J* = 12.6 Hz, 1H), 4.74−4.72 (m, 1H), 4.26 (s, 1H), 3.67 (s, 1H), 3.66−3.56 (m, 1H), 3.56 (s, 3H), 3.55 (s, 3H), 2.98−2.95 (m, 1H), 2.86−2.81 (m, 1H), 2.74−2.62 (m, 3H), 2.28−2.20 (m, 2H), 2.20 (s, 3H), 2.15 (s, 3H), 1.24 (s, 9H), 0.94 (s, 18H), 0.22 (s, 3H), 0.12 (s, 9H); ^{13}C NMR (150 MHz, DMSO-d_6) δ 173.3, 155.6, 155.3, 148.4, 147.3, 146.9, 144.4, 137.2, 132.3, 131.9, 131.6, 130.2, 128.2 (2C), 127.5 (2C), 127.3 (2C), 123.3, 121.8, 118.2, 78.0, 65.1, 63.9, 59.7, 52.3, 50.8, 48.1, 44.5, 38.1, 28.5, 28.0 (2C), 27.9, 27.6, 26.0 (3C), 25.5 (3C), 18.3, 17.9, 16.7, 15.5, −3.8, −4.4, −4.7, −4.8; HRMS calcd for $C_{49}H_{75}BrN_3O_{10}Si_2$ $[M + H]^+$ 1000.4169, found 1000.4182.

Compound 9. To a solution of compound **8** (4.30 g, 4.30 mmol) in CH_2Cl_2 (100 mL) was added Dess−Martin periodinane (3.20 g, 7.52 mmol) and $NaHCO_3$ (5.00 g, 60.20 mmol), and then the mixture was stirred at room temperature for 2 h. The mixture was washed with saturated aq. $NaHCO_3$ and brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (PET/EtOAc = 10:1) afforded the hemiaminal product **9** (4.01 g, 94%) as a white solid. Mp = 112−114 °C; $[\alpha]_D^{20}$ = −75 (*c* 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, DMSO-d_6) δ 7.28−7.19 (m, 5H), 6.97 (s, 1H), 6.70 (s, 1H), 6.68 (s, 1H), 5.74−5.73 (m, 2H), 4.91 (d, *J* = 12.3 Hz, 1H), 4.84 (d, *J* = 12.3 Hz, 1H), 4.68 (dd, *J* = 10.8, 3.6 Hz, 1H), 3.64 (s, 3H), 3.55 (s, 3H), 3.52−3.47 (m, 2H), 3.42−3.31 (m, 3H), 3.15 (t, *J* = 13.2 Hz, 1H), 2.96−2.86 (m, 1H), 2.74 (d, *J* = 13.2 Hz, 1H), 2.28 (s, 3H), 2.17 (s, 3H), 2.07 (s, 9H), 1.00−0.93 (m, 18H), 0.21−0.08 (m, 12H); ^{13}C NMR (150 MHz, DMSO-d_6) δ 167.3, 156.0, 152.1, 148.1, 147.6, 146.9, 144.6, 137.0, 133.7, 131.2, 130.0, 128.2 (2C), 127.6, 127.4 (2C), 123.8, 122.8, 122.0, 119.4, 79.2, 73.5, 65.2, 59.7, 59.5, 57.9, 54.6, 49.2, 44.4, 42.5, 27.4 (3C), 25.9 (3C), 25.5 (3C), 18.3, 17.9, 16.7, 15.7, −4.0, −4.6, −4.7, −4.8; HRMS calcd for $C_{49}H_{76}BrN_4O_{10}Si_2$ $[M + NH_4]^+$ 1015.4278, found 1015.4235.

Compound 10. To an ice-cooled solution of compound **9** (1.90 g, 1.91 mmol) in THF (40 mL) was added TBAF (1.0 M in THF, 5.73 mL), and then the mixture was stirred at 0 °C for 1 h. The reaction was quenched with saturated aq. NH_4Cl and extracted with EtOAc (40 mL × 3). The combined organic phase was washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (PET/EtOAc = 1:1) afforded the deprotected product **10** (1.45 g, 99%) as a white solid. Mp = 84−86 °C; $[\alpha]_D^{20}$ = −85 (*c* 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, DMSO-d_6) δ 9.22 (s, 1H), 9.02 (s, 1H), 7.30−7.16 (m, 5H), 6.88 (s, 1H), 6.76 (s, 1H), 6.52 (s, 1H), 5.74 (s, 1H), 5.64 (s, 1H), 4.88 (s, 2H), 4.66 (d, *J* = 9.0 Hz, 1H), 3.63 (s, 3H), 3.58 (s, 3H), 3.57−3.40 (m, 4H), 3.11−3.03 (m, 2H), 2.89 (t, *J* = 13.2 Hz, 1H), 2.68 (d, *J* = 13.2 Hz, 1H), 2.26 (s, 3H), 2.17 (s, 3H), 0.98 (s, 9H); ^{13}C NMR (150 MHz, DMSO-d_6) δ 167.2, 156.1, 152.2, 148.7, 146.2, 145.1, 144.4, 137.1, 133.7, 131.2, 130.6, 129.3, 128.2 (2C), 127.6, 127.4 (2C), 119.8, 119.4, 118.0, 116.5, 79.0, 73.8, 65.2, 59.8, 59.7, 58.0, 54.9, 48.9, 44.2, 42.2, 30.8, 27.7, 27.3, 16.6, 15.5, 14.0; HRMS calcd for $C_{37}H_{44}BrN_3O_{10}Na$ $[M + Na]^+$ 792.2102, found 792.2071.

Compound 11. A mixture of compound **10** (1.02 g, 1.33 mmol) in TfOH (10 mL) was stirred at room temperature under argon for 2 h, and then the mixture was basified with saturated aq. $NaHCO_3$ and extracted with EtOAc (40 mL × 3). The combined organic phase was washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (CH_2Cl_2/MeOH = 100:2) afforded the pentacyclic product **11** (568 mg, 83%) as a white solid. Mp = 183−185 °C; $[\alpha]_D^{20}$ = −47 (*c* 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, DMSO-d_6) δ 6.46 (s, 1H), 5.23 (d, *J* = 6.6 Hz, 1H), 4.39 (d, *J* = 3.0 Hz, 1H), 3.83 (d, *J* = 12.9 Hz, 1H), 3.74 (s, 1H), 3.64 (s, 3H), 3.61 (s, 3H), 2.96 (d, *J* = 13.2 Hz, 1H), 2.80 (s, 2H), 2.66 (dd, *J* = 11.4, 3.0 Hz, 1H), 2.27 (s, 3H), 2.13 (s, 3H), 2.08 (d, *J* = 13.8 Hz, 1H), 1.95 (dd, *J* = 11.4, 9.6 Hz, 1H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 170.3, 147.5, 146.0, 145.9, 144.5, 130.5, 130.0, 129.7, 129.1, 122.3, 122.2, 119.7, 116.0, 61.1, 60.4, 59.4, 53.4, 51.2, 48.4, 46.6, 36.1, 31.9, 16.5, 15.6; HRMS calcd for $C_{24}H_{29}BrN_3O_5$ $[M + H]^+$ 518.1285, found 518.1274.

Compound 12. To a solution of compound **11** (568 mg, 1.10 mmol) and Et_3N (0.38 mL, 2.75 mmol) in CH_2Cl_2 (60 mL) was added Boc_2O (1.0 M in CH_2Cl_2, 11 mL) dropwise at 0 °C, and then the mixture was stirred at room temperature for 4.5 h. The reaction mixture was washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (CH_2Cl_2/MeOH = 100:1) afforded the N-protected product **12** (616 mg, 91%) as a white solid. Mp = 127−129 °C; $[\alpha]_D^{20}$ = −74 (*c* 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.93 (br s, 2H), 6.46 (s, 1H), 5.52 (s, 1H), 5.50 (s, 1H), 4.39 (s, 1H), 3.81−3.73 (m, 1H), 3.73 (s, 1H), 3.62 (s, 3H), 3.59 (s, 3H), 3.31 (br s, 1H), 3.26 (s, 1H), 2.97 (d, *J* = 13.2 Hz, 1H), 2.90−2.82 (m, 3H), 2.27 (s, 3H), 2.25 (d, *J* = 13.2 Hz, 1H), 2.15 (s, 3H), 1.15 (s, 6H), 0.94 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 170.6, 154.8, 146.3, 146.0, 144.5, 144.4, 132.4, 129.9, 129.8, 129.3, 122.1, 119.9, 119.1, 116.0, 77.4, 61.4, 60.4, 59.8, 53.3, 48.3, 47.4, 44.0, 36.1, 31.8, 27.9 (2C), 27.3, 16.5, 15.4; HRMS calcd for $C_{29}H_{37}BrN_3O_7$ $[M + H]^+$ 618.1809, found 618.1795.

Compound 13. To a solution of compound **12** (507 mg, 0.82 mmol) in CH_3OH (40 mL) were added HCHO (37%, 4.40 mL), $NaBH_3CN$ (520 mg, 8.2 mmol), and acetic acid (7.9 mL), and then the mixture was stirred at room temperature for 2 h. The mixture was concentrated under reduced pressure and then dissolved in EtOAc, washed with saturated aq. $NaHCO_3$ and brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (CH_2Cl_2/MeOH = 100:1) afforded methylated product **13** (516 mg, 99%) as a white solid. Mp = 132−134 °C; $[\alpha]_D^{20}$ = −72 (*c* 0.10, CH_2Cl_2); ^{1}H NMR (300

MHz, DMSO-d_6) δ 9.34 (s, 1H), 8.99 (s, 1H), 6.46 (s, 1H), 5.52 (s, 1H), 5.47 (t, J = 5.1 Hz, 1H), 4.27 (d, J = 2.4 Hz, 1H), 3.84 (d, J = 11.7 Hz, 1H), 3.64 (s, 3H), 3.57 (s, 3H), 3.57−3.54 (m, 1H), 2.99−2.87 (m, 3H), 2.70 (s, 1H), 2.64 (s, 1H), 2.29 (s, 6H), 2.16 (s, 3H), 2.29−2.22 (m, 1H), 1.15 (s, 6H), 0.95 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 170.2, 154.8, 147.0, 146.3, 144.6, 144.4, 132.3, 130.0, 129.3, 128.8, 119.9, 119.4, 119.0, 115.8, 77.4, 60.4, 59.8, 59.2, 58.2, 54.9, 54.6, 47.8, 43.6, 31.3, 30.6, 27.9 (2C), 27.3, 16.5, 15.4; HRMS calcd for $C_{30}H_{39}BrN_3O_7$ $[M + H]^+$ 632.1966, found 632.1971.

Compound 14. To a solution of compound **13** (426 mg, 0.68 mmol) in CH_3OH (40 mL) were added $Pd(OH)_2$ (moist, Pd content 20%, 340 mg) and acetic acid (40 drops); then the mixture was hydrogenated in a Parr apparatus (50 psi H_2) for 10 h at room temperature. The reaction mixture was filtered and concentrated under reduced pressure. The residue was dissolved in EtOAc (50 mL), washed with saturated aq. $NaHCO_3$ and brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (PET/EtOAc = 1:2) afforded hydrogenation product **14** (349 mg, 93%) as a white solid. Mp = 167−169 °C; $[\alpha]_D^{20}$ = −263 (c 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.96 (s, 1H), 8.91 (s, 1H), 6.45 (s, 2H), 5.50 (t, J = 5.4 Hz, 1H), 5.32 (s, 1H), 4.18 (s, 1H), 3.83 (d, J = 12.6 Hz, 1H), 3.62 (s, 3H), 3.60 (s, 3H), 3.50 (d, J = 6.0 Hz, 1H), 3.10 (dd, J = 17.1, 6.6 Hz, 1H), 2.96−2.86 (m, 2H), 2.63 (s, 1H), 2.57 (s, 1H), 2.31 (s, 3H), 2.31−2.25 (m, 1H), 2.17 (s, 3H), 2.16 (s, 3H), 1.17 (s, 6H), 0.95 (s, 3H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 170.6, 154.7, 147.3, 146.3, 144.3, 143.9, 132.4, 129.4, 129.3, 128.7, 120.6, 119.9, 119.2, 117.2, 77.4, 59.9, 59.8, 59.2, 58.3, 54.9, 54.6, 47.4, 43.8, 31.4, 28.1, 27.9 (2C), 27.3, 15.6, 15.4; HRMS calcd for $C_{30}H_{40}N_3O_7$ $[M+H]^+$ 554.2861, found 554.2871.

Compound 15. To a stirred solution of compound **14** (94 mg, 0.17 mmol) in THF (15 mL) was added $LiAlH_4$ (32 mg, 0.84 mmol) at −17 °C under argon, and then the mixture was stirred for 2 h at room temperature. The reaction was cooled to −17 °C, which was followed by careful addition of KCN (88.40 mg, 1.36 mmol) in 2.7 mL of H_2O and acetic acid (0.2 mL, 3.40 mmol). The mixture was stirred for 10 h at room temperature. The reaction was then quenched by saturated aq. $NaHCO_3$ and extracted with EtOAc (15 mL × 3). The combined organic phase was washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (CH_2Cl_2/MeOH = 100:1) afforded cyanided product **15** (79 mg, 82%) as a white solid. Mp = 90−92 °C; $[\alpha]_D^{20}$ = +29 (c 0.10, CH_2Cl_2); ^{1}H NMR (600 MHz, DMSO-d_6) δ 8.78 (s, 1H), 8.57 (s, 1H), 6.35 (s, 1H), 6.26 (s, 1H), 4.53 (s, 1H), 4.32 (s, 1H), 3.98 (s, 1H), 3.86 (s, 1H), 3.58 (s, 3H), 3.55 (s, 3H), 3.23 (d, J = 7.2 Hz, 1H), 3.19 (t, J = 9.0 Hz, 1H), 2.99 (d, J = 12.0 Hz, 1H), 2.92 (dd, J = 18.0, 8.4 Hz, 1H), 2.89 (d, J = 7.2 Hz, 1H), 2.66 (d, J = 12.0 Hz, 1H), 2.44 (d, J = 18.0 Hz, 1H), 2.40 (dd, J = 16.2, 8.4 Hz, 1H), 2.16 (s, 3H), 2.14 (s, 3H), 2.09 (s, 3H), 1.10 (s, 9H); ^{13}C NMR (150 MHz, DMSO-d_6) δ 177.8, 154.9, 147.3, 145.9, 143.8, 143.3, 131.4, 130.4, 128.6, 128.4, 119.4 (2C), 118.9, 117.6, 77.2, 68.2, 65.7, 59.9, 59.4, 56.8, 56.1, 54.5, 43.4, 41.2, 31.4, 27.9, 27.4, 24.9, 21.7, 15.6, 15.4; HRMS calcd for $C_{31}H_{41}N_4O_6$ $[M + H]^+$ 565.3021, found 565.3027.

Compound 16. A mixture of compound **15** (60 mg, 0.106 mmol) in TFA (1 mL) was stirred at room temperature for 1 h, and then the mixture was basified with saturated aq. $NaHCO_3$ and extracted with EtOAc (15 mL × 3). The combined organic phase was washed with brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (CH_2Cl_2/MeOH = 30:1) afforded deprotected product **16** (44 mg, 90%) as a white solid. Mp = 140−142 °C; $[\alpha]_D^{20}$ = +40 (c 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.45 (s, 1H), 6.39 (s, 1H), 4.72 (s, 2H), 4.10 (br s, 1H), 4.01 (s, 1H), 3.98 (d, J = 2.1 Hz, 1H), 3.75 (s, 3H), 3.72 (s, 3H), 3.34 (d, J = 7.8 Hz, 1H), 3.30 (d, J = 12.0 Hz, 1H), 3.06 (dd, J = 18.0, 8.1 Hz, 1H), 2.84 (dd, J = 13.5, 1.8 Hz, 1H), 2.75 (dd, J = 15.0, 2.1 Hz, 1H), 2.58 (dd, J = 13.5, 5.4 Hz, 1H), 2.47 (d, J = 18.0 Hz, 1H), 2.32 (s, 3H), 2.23 (s, 3H), 2.21 (s, 3H), 2.06 (dd, J = 15.0, 12.3 Hz, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 146.6, 145.9, 144.4, 142.7, 131.7, 130.8, 129.1, 128.7, 120.8(2C), 119.5, 118.2, 116.9, 60.7, 60.4, 60.3, 59.1, 56.6, 55.4, 45.5, 41.8, 32.1, 29.7, 25.8, 15.7, 15.6; HRMS calcd for $C_{26}H_{33}N_4O_4$ $[M + H]^+$ 465.2496, found 465.2508.

Compound 17. To a solution of compound **16** (40 mg, 0.086 mmol) in CH_2Cl_2 (5 mL) were added pyruvic acid (10.5 mg, 0.129 mmol), DMAP (10.5 mg, 0.086 mmol), and EDC (25.2 mg, 0.129 mmol), and then the mixture was stirred at room temperature for 10 h. The mixture was diluted with CH_2Cl_2, washed with saturated aq. $NaHCO_3$ and brine, dried over anhydrous Na_2SO_4, and concentrated under reduced pressure to give the crude product. Purification by column chromatography (PET/EtOAc = 1:1) afforded pyruvamide **17** (37 mg, 80%) as a white solid. Mp = 100−102 °C; $[\alpha]_D^{20}$ = +4 (c 0.10, CH_2Cl_2); ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.46 (s, 1H), 6.44 (s, 1H), 6.40 (br s, 1H), 5.82 (s, 1H), 5.70 (s, 1H), 4.20 (br s, 1H), 4.09 (s, 1H), 4.01 (d, J = 1.8 Hz, 1H), 3.80 (s, 3H), 3.76 (s, 3H), 3.58−3.49 (m, 1H), 3.45−3.38 (m, 1H), 3.35 (d, J = 7.8 Hz, 1H), 3.30−3.29 (m, 1H), 3.02 (dd, J = 18.3, 8.1 Hz, 1H), 2.78 (d, J = 17.7 Hz, 1H), 2.44 (d, J = 17.7 Hz, 1H), 2.30 (s, 3H), 2.24 (s, 3H), 2.23 (s, 3H), 2.22 (s, 3H), 2.10 (dd, J = 16.2, 11.4 Hz, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 196.1, 159.9, 146.6, 144.9, 143.6, 129.3, 121.3 (2C), 120.9 (2C), 117.6, 60.8, 59.9, 56.7, 56.6, 55.4, 41.6, 41.3, 31.8, 29.7, 25.5, 24.4, 22.7, 15.9, 15.7; HRMS calcd for $C_{29}H_{35}N_4O_6$ $[M + H]^+$ 535.2551, found 535.2555.

(−)-Saframycin A (1). To a solution of compound **17** (12 mg, 0.022 mmol) in CH_3CN (2 mL) were added salcomine (7.5 mg, 0.022 mmol) at room temperature, and the dark suspension was stirred in air for 2 h. The mixture was purified by column chromatography (PET/EtOAc = 1:1) and pTLC (PET/EtOAc = 1:1) to give (−)-saframycin A (10.6 mg, 87%) as a yellow solid. Mp = 120−122 °C; $[\alpha]_D^{20}$ = −98 (c 0.10, CH_2Cl_2); $[\alpha]_D^{20}$ = +18 (c 0.10, MeOH); ^{1}H NMR (600 MHz, $CDCl_3$) δ 6.67 (br dd, J = 8.1, 4.2 Hz, 1H), 4.06 (br d, J = 0.6 Hz, 1H), 4.03 (s, 3H), 4.02 (s, 3H), 3.99 (d, J = 2.4 Hz, 1H), 3.97 (m, 1H), 3.73 (ddd, J = 13.8, 9.0, 1.5 Hz, 1H), 3.43 (br d, J = 7.2 Hz, 1H), 3.26 (dt, J = 14.4, 4.2 Hz, 1H), 3.13 (dt, J = 11.4, 3.0 Hz, 1H), 2.88 (dd, J = 18.0, 3.0 Hz, 1H), 2.82 (dd, J = 21.0, 7.8 Hz, 1H), 2.31 (s, 3H), 2.26 (s, 3H), 2.24 (d, J = 21.0 Hz, 1H), 1.99 (s, 3H), 1.92 (s, 3H), 1.28 (ddd, J = 18.0, 11.4, 3.0 Hz, 1H); ^{13}C NMR (150 MHz, $CDCl_3$) δ 196.7, 186.6, 185.3, 182.4, 180.8, 160.1, 155.9, 155.6, 141.5, 141.2, 135.5 (2C), 129.2, 128.3, 116.6, 61.1, 60.9, 58.2, 56.2, 54.4, 54.1, 53.9, 41.6, 40.6, 25.0, 24.3, 21.5, 8.7 (2C); HRMS calcd for $C_{29}H_{31}N_4O_8$ $[M + H]^+$ 563.2136, found 563.2148.

■ ASSOCIATED CONTENT

Supporting Information. Full experimental details and copies of NMR spectral data. This material is available free of charge via the Internet at http://pubs.acs.org.

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REFERENCES

(1) Arai, T.; Takahashi, K.; Kubo, A. *J. Antibiot.* **1977**, *30*, 1015–1018.

(2) Arai, T.; Takahashi, K.; Nakahara, S.; Kubo, A. *Experientia* **1980**, *36*, 1025–1027.

(3) Scott, J. D.; Williams, R. M. *Chem. Rev.* **2002**, *102*, 1669–1730.

(4) Molinski, T. F.; Dalisay, D. S.; Lievens, S. L.; Saludes, J. P. *Nat. Rev. Drug Discovery* **2009**, *8*, 69–85.

(5) Ocio, E. M.; Maiso, P.; Chen, X.; Garayoa, M.; Alvarez-Fernandez, S.; San-Segundo, L.; Vilanova, D.; Lopez-Corral, L.; Montero, J. C.; Hernandez-Iglesias, T.; de Alava, E.; Galmarinini, C.; Aviles, P.; Cuevas, C.; San-Miguel, J. F.; Pandiella, A. *Blood* **2009**, *113*, 3781–3791.

(6) Leal, J. F. M.; Garcia-Hernandez, V.; Moneo, V.; Domingo, A.; Bueren-Calabuig, J. A.; Negri, A.; Gago, F.; Guillen-Navarro, M. J.; Aviles, P.; Cuevas, C.; Garcia-Fernandez, L. F.; Galmarini, C. M. *Biochem. Pharmacol.* **2009**, *78*, 162–170.

(7) Fukuyama, T.; Yang, L.; Ajeck, K. L.; Sachleben, R. A. *J. Am. Chem. Soc.* **1990**, *112*, 3712–3713.

(8) Martinez, E. J.; Corey, E. J. *Org. Lett.* **1999**, *1*, 75–77.

(9) Myers, A. G.; Kung, D. W.; Zhong, B.; Movassaghi, M.; Kwon, S. *J. Am. Chem. Soc.* **1999**, *121*, 8401–8402.

(10) Myers, A. G.; Schnider, P.; Kwon, S.; Kung, D. W. *J. Org. Chem.* **1999**, *64*, 3322–3327.

(11) Myers, A. G.; Kung, D. W. *J. Am. Chem. Soc.* **1999**, *121*, 10828–10829.

(12) Tang, Y. F.; Liu, Z. Z.; Chen, S. Z. *Tetrahedron Lett.* **2003**, *44*, 7091–7094.

(13) Liu, Z. Z.; Wang, Y.; Tang, Y. F.; Chen, S. Z.; Chen, X. G.; Li, H. Y. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 1282–1285.

(14) Liao, X. W.; Liu, W.; Dong, W. F.; Guan, B. H.; Chen, S. Z.; Liu, Z. Z. *Tetrahedron* **2009**, *65*, 5709–5715.

(15) Liao, X. W.; Dong, W. F.; Liu, W.; Guan, B. H.; Liu, Z. Z. *J. Heterocycl. Chem.* **2010**, *47*, 50–53.

(16) Liu, W.; Dong, W. F.; Liao, X. W.; Yan, Z.; Guan, B. H.; Wang, N.; Liu, Z. Z. *Bioorg. Med. Chem. Lett.* **2011**, *21*, 1419–1421.

(17) Hansen, D. W.; Pilipauskas, D. *J. Org. Chem.* **1985**, *50*, 945–950.

(18) Colucci, W. G.; Tung, R. D.; Petri, J. A.; Rich, D. H. *J. Org. Chem.* **1990**, *55*, 2895–2903.

(19) Kende, A. S.; Liu, K.; Kaldor, I.; Dorey, G.; Koch, K. *J. Am. Chem. Soc.* **1995**, *117*, 8258–8270.

(20) Dess, P. B.; Martin, J. C. *J. Org. Chem.* **1983**, *48*, 4155–4156.

(21) Yokoyama, A.; Ohwada, T.; Shudo, K. *J. Org. Chem.* **1999**, *64*, 611–617.

(22) Lane, J. W.; Chen, Y.; Williams, R. M. *J. Am. Chem. Soc.* **2005**, *127*, 12684–12690.

(23) Zhou, B. S.; Guo, J. S.; Danishefsky, S. J. *Org. Lett.* **2002**, *4*, 43–46.

(24) Saito, N.; Obara, Y.; Aihara, T.; Harada, S.; Shida, Y.; Kubo, A. *Tetrahedron* **1994**, *50*, 3915–3928.