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# Practical Large-Scale Synthesis of Endothelin Receptor Antagonist S-0139

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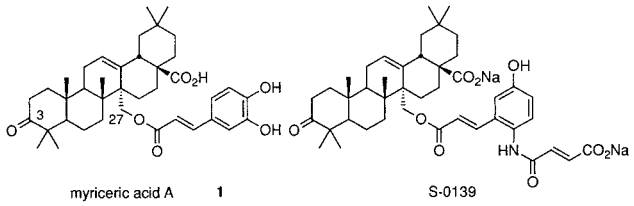
**Abstract:**

**Semisynthetic endothelin receptor antagonist S-0139 was synthesized in 14 steps from oleanolic acid 2 in a 20% overall yield on a multi-kilogram scale. Our previous synthesis of the oleanane skeleton was modified and improved to give phosphonate 9 as a key intermediate. The side chain on the 27-position was introduced by Horner−Wadsworth−Emmons olefination of phosphonate 9 with aldehyde 5. Aldehyde 5 was prepared in a one-pot reduction−acylation process starting from 5-hydroxy-2-nitrobenzaldehyde. The entire sequence of the synthesis can be done without chromatography and yields S-0139 of high purity.**

## Introduction

Myriceric acid A **1**[1] is a potent endothelin receptor antagonist[2] that was found by screening the Shionogi natural product library and that was isolated from a crude extract of twigs of the southern bayberry, *Myrica cerifera*. It is an oleanane triterpene which is characterized by 3-oxo and 27-caffeoyloxy groups. An extensive modification study of the 27-acyloxy group led to S-0139 (formerly 97-139),[3] which is a specific antagonist of endothelin A receptor and currently in clinical study. The characteristics of S-0139 reside in its enhanced potency and water solubility brought by the carboxyl group of the side chain. We previously reported the practical synthesis of myriceric acid A, **1**,[4] from naturally abundant oleanolic acid **2** by way of Horner−Wadsworth−Emmons (HWE)[5] phosphonate **4** as a key intermediate. On the basis of Barton's method,[6] we optimized each step including the Barton reaction and developed an efficient method that resolved most of the problems of Barton's original method.




We employed the HWE olefination of phosphonate **4** with aldehyde **5** giving **6** (Scheme 1) to supply S-0139 at the early stage of pharmacological evaluation. Although the overall conversion itself was efficient (26% from oleanolic acid to S-0139), it had two major problems. One was chromatographic purification of the HWE olefination product **6**. The olefination gave three other byproducts, and careful silica gel chromatography was necessary to avoid the contamination of the final S-0139 which will cause a serious concern for the New Drug Application. These byproducts were assumed to be derived from aldol condensation of **6** and aldehyde **5**. The other problem was inefficient synthesis of the side chain aldehyde **5**. There had been no efficient method because of the instability of 2-amino-5-hydroxybenzaldehyde, **15**. Here, we describe the improved procedure for the synthesis of S-0139, in which we changed the sequence of reactions and overcame the difficulties of the previous method. We were able to establish the large-scale synthesis of S-0139 of high purity which satisfied the specifications of a bulk pharmaceutical chemical.

## Results and Discussion

**Original Procedure.** HWE olefination of phosphonate **4**[4] worked well for a number of aldehydes, and 27-*O*-cinnamoylmyricerone derivatives were obtained in high yields (Scheme 1, upper). For the supply of S-0139 in the early stages, this method was adopted on a scale of several hundred grams. However, HWE condensation product **6** was contaminated by three byproducts, which were detected by HPLC and found to be two diastereomeric aldol products **12a** and **12b** and their dehydrated $\alpha,\beta$-unsaturated ketone **13**.

Under proper reaction conditions, the yields of **6**, **12**, and **13** were 92.7, 0.6, and 5.3%, respectively; however, they changed to 65.1, 4.3, and 21.7%, respectively, with prolonged reaction time. Clearly, these byproducts were generated under the basic condition of the HWE olefination. The acidic hydrogen on the C-2 was deprotonated to give enolate, which

(1) For isolation of myriceric acid A, see: Sakurawi, K.; Yasuda, F.; Tozyo, T.; Nakamura, M.; Sato, T.; Kikuchi, J.; Terui, Y.; Ikenishi, Y.; Iwata, T.; Takahashi, K.; Konoike, T.; Mihara, S.; Fujimoto, M. *Chem. Pharm. Bull.* **1996**, *44*, 343. For biological study, see: Fujimoto, M.; Mihara, S.; Nakajima, S.; Ueda, M.; Nakamura, M.; Sakurai, K. *FEBS Lett.* **1992**, *305*, 41. Mihara, S.; Fujimoto, M. *Eur. J. Pharmacol.* **1993**, *246*, 33. Mihara, S.; Sakurai, K.; Nakamura, M.; Konoike, T.; Fujimoto, M. *Eur. J. Pharmacol.* **1993**, *247*, 219.

(2) For endothelins and their receptor antagonists, see: Cheng, X.-M.; Nikam, S. S.; Doherty, A. M. *Curr. Med. Chem.* **1994**, *1*, 271.

(3) Mihara, S.; Nakajima, S.; Matsumura, S.; Konoike, T.; Fujimoto, M. *J. Pharmacol. Exp. Ther.* **1994**, *268*, 1122.

(4) Konoike, T.; Takahashi, K.; Araki, Y.; Horibe, I. *J. Org. Chem.* **1997**, *62*, 960.

(5) For a review, see: Maryanoff, B. E.; Reitz, A. B. *Chem. Rev.* **1989**, *89*, 863.

(6) Barton, D. H. R.; Sammes, P. G.; Silva, M. *Tetrahedron Suppl.* **1966**, 57. See also references in our previous paper.[4]

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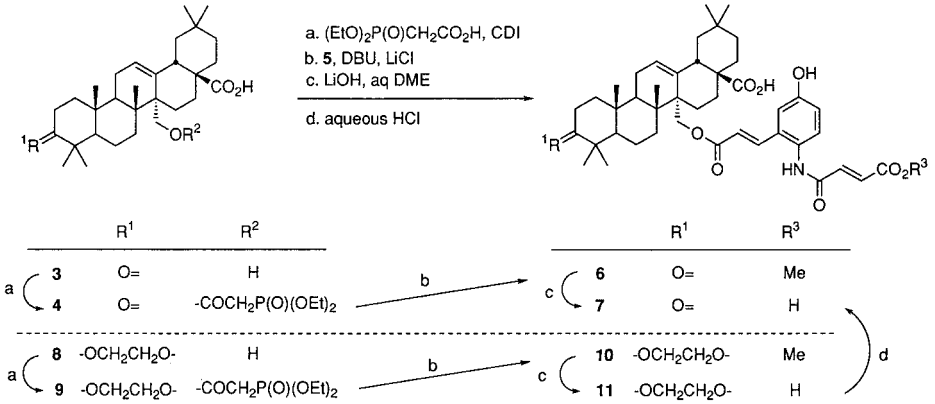
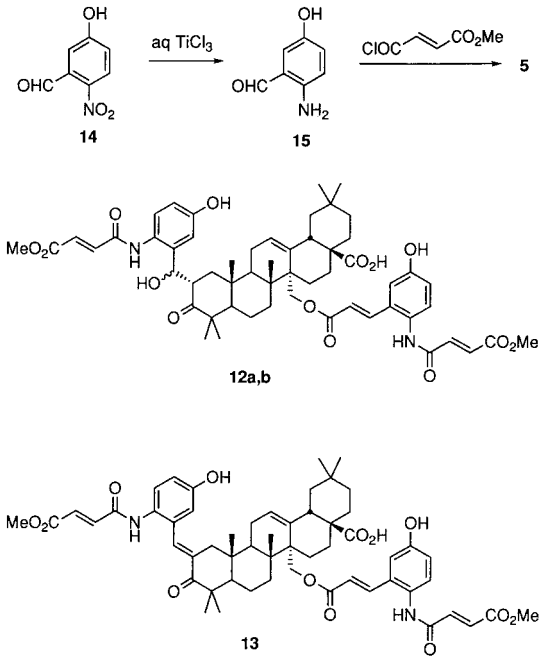
**Scheme 1**




**Scheme 2**




reacted with aldehyde **5** to give aldol products **12a**,**b** and α,β-unsaturated ketone **13**. Purification of the desired **6** required silica gel chromatography to remove the impurities, and this was a major difficulty in the large-scale synthesis. Several attempts for purifying the intermediate **6** and free acid **7** of S-0139 by crystallization were unsuccessful. Acid **7** was obtained as crystalline needles, but we were unable to obtain S-0139 pure enough for clinical evaluations, even after several recrystallization steps.

**Improved Procedure.** To circumvent the problem, we modified the sequence of reactions and changed the order of synthetic steps (Scheme 1, lower). Instead of using **3** for phosphonate formation, we used myricerone ketal **8**, which itself is a precursor of myricerone **3**. The hydrogen at C-2 of myricerone ketal **8** is not acidic, and it is devoid of enolate formation under basic conditions. Myricerone ketal **8** was first acylated with diethylphosphonoacetic acid by means of 1,1′-carbonyldiimidazole to give 27-*O*-diethylphosphonoacetylmyricerone ketal **9**. This is a crystalline compound which can be isolated in a pure form by simple crystallization. This phosphonate was condensed with aldehyde **5** to efficiently and quantitatively give α,β-unsaturated ester **10**, and neither aldols nor an unsaturated ketone was detected as a byproduct in **10**. Ester **10** could be purified by crystallization; however, further investigation of the following steps proved that purification of **10** by crystallization at this step could be eliminated. The crude extract of **10** was treated with LiOH to hydrolyze the methyl ester, and ketal acid **11** was obtained in quantitative yield. Ketal acid **11** was treated with aqueous HCl to deprotect the ketal group, and free acid **7** of S-0139 was obtained. Deketalization was an equilibrium reaction, and starting ketal **11** was not converted completely to free acid **7** of S-0139. Acid **7** obtained after workup contained starting material **11** (ca 0.3% by HPLC), which is the only major impurity in **7**. The desired acid **7** was crystallized from EtOAc/MeCN, and the resulting material contained less than 0.1% of precursor **11**. Crystalline free acid **7** of S-0139 was treated with 2 equiv of aqueous NaOH and then freeze-dried to give an amorphous pale yellow powder of disodium salt, S-0139.

**Synthesis of Side Chain Aldehyde.** Side chain aldehyde **5** was another requisite building block for S-0139, and we envisioned its preparation from 2-amino-5-hydroxybenzaldehyde, **15**, and *trans*-β-methoxycarbonylacryloyl chloride.[7] A survey of the literature revealed no reliable and practical procedure for **15** although its protected forms have been reported recently as an intermediate for the synthesis of camptothecin analogues.[8] Initially, aminohydroxybenzaldehyde **15** was prepared by photoreaction of indazole;[9] however, the photoreaction was an obvious drawback for the large-scale preparation. From these initial experiments, we concluded that **15** had not been available presumably owing to its lability. Apparently, reduction of commercially available 5-hydroxy-2-nitrobenzaldehyde, **14**,[10] was the most straightforward way to **15**. However, there had been no report on the direct reduction of **14**. Consequently, we devised an efficient alternative method to reduce the nitro group to an

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(9) Georgarakis, E.; Schmid, H.; Hansen, H.-J. *Helv. Chim. Acta* **1979**, *62*, 234.
(10) For synthesis of **14**, see: Shaw, K. N. F.; Gudmundson, A.; Morris, A. G. *Biochem. Prepr*. **1962**, *9*, 12. For synthetic application, see: Shaw, K. N. F.; Morris, A. J. *Biochem. Prepr*. **1962**, *9*, 92. Fürstner, A.; Jumbam, D. N.; Seidel, G. *Chem. Ber*. **1994**, *127*, 1125.

amino group. We tried a number of reducing reagents, and Somei's reduction by aqueous $TiCl_3$[11] was found suitable for preparation of **15** because of its low temperature, weak acidic condition, and rapid reaction. We finally devised a two-step, one-pot procedure for the side chain aldehyde **5**. Hydroxynitrobenzaldehyde **14** was reduced by aqueous $TiCl_3$, and aminoaldehyde **15**, generated in situ, was treated with *trans*-$\beta$-methoxycarbonylacryloyl chloride in the presence of sodium acetate. On completion of the acylation, desired aldehyde **5** precipitated and was collected by filtration (62% yield). Aldehyde **5** thus obtained was used for HWE olefination with phosphonate **9** as described in the preceding section.

In conclusion, we developed an efficient and practical partial synthesis of S-0139 from oleanolic acid **2**, which has been carried out on a multi-kilogram scale for preparation of S-0139 for clinical evaluation.

## Experimental Section

**General.** Reactions were carried out under a nitrogen atmosphere. Solvent removal was accomplished under reduced pressure. TLC was performed with Merck precoated TLC plates silica gel 60 $F_{254}$, and compound visualization was effected with 10% $H_2SO_4$ containing 5% ammonium molybdate and 0.2% ceric sulfate. Chromatography was done with Merck silica gel 60 (70−230 mesh). HPLC analysis was performed under the following conditions: column, Cosmosil 5C18 AR 4.6 mm × 150 mm; guard column, Lichrospher 100 RP 18; solvent, MeCN/$H_2O$/AcOH; flow rate, 1 mL/min; detection, 197 nm. Melting points are uncorrected. $^1$H NMR and $^{13}$C NMR spectra were determined as $CDCl_3$ solutions at 200 and 50.3 MHz. *J* values are given in hertz. High-resolution mass spectra (HR-LSIMS) were recorded on a HITACHI M-90 instrument.

**Starting Materials.** Starting materials, myricerone **3**, 27-*O*-diethylphosphonoacetylmyricerone, **4**, and myricerone ketal **8** were prepared by our method reported previously.[4] Other compounds were commercial products or prepared by known methods.

**5-Hydroxy-2-(3-methoxycarbonylacryloylamino)benzaldehyde (5).** To a suspension of 5-hydroxy-2-nitrobenzaldehyde (225 g, 1.35 mol) in AcOH (675 mL) was added 20% aqueous $TiCl_3$ (4.69 L) over 45 min with stirring at 25 °C, and the resulting solution was stirred at room temperature for 1.5 h. To the solution were added portionwise NaOAc (666 g) and EtOAc (225 mL) under ice−salt cooling temperature. Next, a solution of $\beta$-methoxycarbonylacryloyl chloride (1189 g, 6.75 mol) in EtOAc (1125 mL) was added dropwise over 60 min at 22−25 °C, and stirring was continued at 0 °C for 2 h. The resulting precipitates were collected by filtration and washed with $H_2O$ (3.5 L) to give crude **5** contaminated with hydrogen methyl fumarate. The obtained precipitates were suspended in aqueous $NaHCO_3$ (750 g in 10 L of $H_2O$), and then collected by filtration. The precipitate was washed with $H_2O$ (2 L) and dried to give crude **5** (300 g). Aldehyde **5** was dissolved in methyl ethyl ketone (12 L) on heating at 65 °C and treated with charcoal. Charcoal and insoluble materials were removed by filtration, and the filtrate was condensed to 2.0 kg and kept at 0 °C for 12 h. The resulting slurry was filtered, and the precipitate was collected to give **6** as a yellow crystalline powder (208 g, 62%): mp 215−216 °C; TLC $R_f$ 0.34 (toluene/EtOAc = 2/1); HPLC; IR (Nujol) 3394, 3206, 1738, 1670, 1163 cm$^{-1}$; $^1$H NMR ($d_6$-DMSO) $\delta$ 3.77 (s, 3), 6.73 (d, 1, $J$ = 15.5), 7.10 (dd, 1, $J$ = 2.9, 8.6), 7.20 (d, 1, $J$ = 15.3), 7.21 (d, 1, $J$ = 2.9), 7.65 (d, 1, $J$ = 8.6), 9.92 (s, 1), 9.94 (s, 1), 10.7 (s, 1); $^{13}$C NMR ($d_6$-DMSO) $\delta$ 52.0, 115.4, 121.9, 125.4, 128.2, 129.3, 130.5, 137.3, 155.0, 162.2, 165.3, 191.7. Anal. Calcd for $C_{12}H_{11}NO_5$: C, 57.83; H, 4.45; N, 5.62. Found: C, 57.83; H, 4.38; N, 5.77.

**Methyl Ester of S-0139 (6).** A solution of **4** (145.7 g, 0.203 mol) and aldehyde **5** (60.8 g, 0.244 mol) in DMF (1.06 L) was cooled in an ice-bath, and to the solution were added DBU (75.9 mL, 0.508 mol) and LiCl (21.5 g, 0.507 mol). The mixture was stirred for 7.5 h at room temperature and then poured into a mixture of EtOAc (10.7 L), 2 N HCl (0.72 L) and ice−water (1.46 L). The organic layer was separated, washed with $H_2O$ (1.45 L ×2), and then condensed to give crude **6**. Pure sample **6** was obtained by silica gel chromatography (85%) to remove byproducts **12** and **13**. **6**: HPLC (MeCN/$H_2O$/AcOH (60/40/0.1)) $t_R$ 5.4 min; IR (KBr) 3306, 2936, 1691, 1634 cm$^{-1}$; $^1$H NMR $\delta$ 0.83 (s, 3), 0.84 (s, 3), 0.93 (s, 3), 1.03 (s, 3), 1.04 (s, 3), 1.07 (s, 1), 1.0−2.1 (m, 20), 2.2−2.8 (m, 2), 2.8−3.0 (m, 1), 3.84 (s, 3), 4.16 (d, 2, $J$ = 13.0), 4.40 (d, 2, $J$ = 13.0), 5.60 (br s, 1), 6.27 (d, 2, $J$ = 16.0), 6.90 (dd, 1, $J$ = 8.8, 2.8), 6.94 (d, 1, $J$ = 15.2), 7.07 (d, 1, $J$ = 2.8), 7.21 (d, 1, $J$ = 15.2), 7.44 (d, 1, $J$ = 8.8), 7.74 (d, 1, $J$ = 16.0); $^{13}$C NMR $\delta$ 15.8, 18.6, 20.7, 21.9, 23.9, 24.0, 24.5, 25.0, 27.1, 31.6, 33.5, 33.6, 34.0, 34.8, 34.9, 38.0, 40.2, 41.1, 42.7, 46.1, 46.8, 47.4, 48.4, 52.8, 56.3, 66.7, 113.6, 119.3, 120.8, 128.0, 128.7, 129.8, 131.8, 133.0, 137.6, 138.8, 141.5, 158.1, 165.3, 167.1, 168.0, 181.5, 220.3. HR-LSIMS $m/z$ 744.4103 $[M + H]^+$ (calcd for $C_{44}H_{58}NO_9$, 744.4108).

**Byproducts 12a,b and 13.** Starting material **4** (1 g, 1.40 mmol) was treated with aldehyde **5** for 40 h under conditions similar to those described above. HPLC of the reaction mixture showed the ratio of **6**, **12**, and **13** was 65.1, 4.3, and 21.7%, respectively. After a usual workup, the three byproducts were separated by silica gel chromatography and HPLC. **12a**: TLC $R_f$ 0.47 (EtOAc/AcOH/$H_2O$ = 60/1/1). HPLC (MeCN/$H_2O$/AcOH (60/40/0.1)) $t_R$ 3.3 min; $^1$H NMR ($CDCl_3$ + $CD_3OD$) $\delta$ 0.68 (s, 3), 0.74 (s, 3), 0.77 (s, 3), 0.89 (s, 3), 1.06 (s, 3), 1.15 (s, 3), 1.0−1.9 (m, 20), 2.75−2.90 (m, 1), 3.04 (dt, 1, $J$ = 10.0, 10.0), 3.86 (s, 3), 3.88 (s, 3), 3.90 (d, 1, $J$ = 12.3), 4.43 (d, 1, $J$ = 12.3), 4.82 (d, 1, $J$ = 9.2), 5.51 (s, 1), 5.75 (dd, 1, $J$ = 8.7, 2.9), 6.08 (d, 1, $J$ = 15.8), 6.65 (d, 1, $J$ = 2.6), 6.9−7.6 (m, 9). LSIMS $m/z$ 993 $[M + H]^+$. **12b**: HPLC (MeCN/$H_2O$/AcOH (60/40/0.1)) $t_R$ 3.9 min; $^1$H NMR ($CDCl_3$+$CD_3OD$) $\delta$ 0.7−2.0 (m, 38), 2.8−3.1 (m 2), 3.86 (s, 3), 3.87 (s, 3), 4.01 (d, 2, $J$ = 12.7), 4.38 (d, 2, $J$ = 12.7), 5.17 (d, 1, $J$ = 4.1), 5.55 (s, 1), 6.04 (dd, 1, $J$ = 8.8, 2.9), 6.17 (d, 1, $J$ = 15.9), 6.50 (d, 1, $J$ = 2.8), 6.6−7.8 (m, 9). LSIMS $m/z$ 993 $[M + H]^+$. **13**: HPLC (MeCN/$H_2O$/AcOH (60/40/0.1)) $t_R$ 4.2 min; $^1$H NMR ($CDCl_3$ + $CD_3$-

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OD) $\delta$ 0.78 (s, 3), 0.79 (s, 3), 0.82 (s, 3), 0.89 (s, 3), 1.14 (s, 6), 1.0−2.0 (m, 20), 2.59 (d, 1, $J$ = 16.7), 2.8−2.9 (m, 1), 3.81 (s, 3), 3.82 (s, 3), 4.04 (d, 2, $J$ = 13.3), 4.36 (d, 1, $J$ = 13.3), 5.50 (s, 1), 6.25 (d, 1, $J$ = 15.9), 6.51 (dd, 1, $J$ = 8.8, 2.8), 6.64 (d, 1, $J$ = 2.8), 6.83 (d, 1, $J$ = 15.4), 6.93 (d, 1, $J$ = 15.5), 6.95 (dd, 1, $J$ = 8.8, 2.8), 7.07 (d, 1, $J$ = 2.8), 7.1−7.4 (m, 5), 7.39 (d, 1, $J$ = 8.8), 7.56 (d, 1, $J$ = 16.0). LSIMS $m/z$ 975 $[M + H]^+$.

**Ketal Phosphonate 9.** Myricerone ketal **8** was prepared by a previously reported method[4] (11.88 mol scale) as a solution in EtOAc (24 L). To the solution was added diethylphosphonoacetic acid (5.82 kg, 29.7 mol), and the resulting mixture was concentrated under reduced pressure to remove water. The residue was diluted by $CH_2Cl_2$ (35 L), and 1,1′-carbonyldiimidazole (4.98 kg, 29.7 mol) was added portionwise over 10 min to the solution. After the mixture was stirred at 45 °C for 3 h, 3.5% aqueous HCl (86 kg) was added, the resulting organic layer was separated, and the aqueous layer was extracted with $CH_2Cl_2$ (40 L). Each organic layer was washed with water (86 kg), combined, and concentrated to 21 L. EtOAc (105 L) was added to the residue, and the mixture was stirred for 30 min to give a slurry of **9**, which was further concentrated to 71 L. The precipitates were collected by filtration and washed with cold EtOAc (35 L) to give **9** (5.75 kg, 70%) as a colorless crystalline powder: mp 215−217 °C; TLC $R_f$ 0.79 (toluene/EtOAc =1/1); IR ($CHCl_3$) 2991, 1730, 1695, 1470, 1388, 1262 $cm^{-1}$; $^1H$ NMR $\delta$ 0.73 (s, 3), 0.82 (s, 3), 0.89 (s, 3), 0.90 (s, 3), 0.93 (s, 6), 0.95 (s, 3), 1.1−2.0 (m, 25), 2.8−2.95 (m, 1), 2.96 (d, 2, $J$ = 21.7), 3.8−4.0 (m, 4), 4.05−4.36 (m, 6), 5.56 (br s, 1); $^{13}C$ NMR $\delta$ 15.5, 16.4, 18.1, 18.3, 20.1, 22.7, 22.7, 22.9, 23.5, 23.6, 26.7, 30.6, 32.4, 32.9, 33.1, 33.6, 34.5, 36.8, 37.1, 39.9, 40.5, 42.0, 45.0, 45.1, 46.2, 48.2, 53.2, 62.7, 62.8, 64.7, 64.8, 66.9, 112.7, 127.5, 137.2, 165.6, 183.8. $[\alpha]^{24}_D$ +50.5° ($c$ 1.004, $CHCl_3$). Anal. Calcd for $C_{38}H_{61}O_9P$: C, 65.87; H, 8.87; P, 4.47. Found: C, 65.72; H, 8.76; P, 4.68. HR-LSIMS $m/z$ 715.3953 $[M + Na]^+$ (calcd for $C_{38}H_{61}O_9NaP$, 715.3948).

**Ketal Methyl Ester 10.** To a solution of ketal phosphonate **9** (3.2 kg, 4.62 mol), aldehyde **5** (1.27 kg, 5.08 mol), and LiCl (0.49 kg, 11.6 mol) in DMF (13 L) was added DBU (1.76 kg, 11.6 mol) at 10 °C, and the mixture was stirred at 20 °C for 6 h and left without stirring overnight. EtOAc (32 L), water (6.4 kg), and 3.5% HCl (33 kg) were added to the mixture, and the organic layer was separated. The aqueous layer was extracted with EtOAc (32 L). Organic layers were washed three times with 20% aqueous NaCl, combined, and then concentrated to 16 L. Dimethoxyethane (48 L) was added to the residue, and the resulting solution was concentrated under reduced pressure to 16 L to remove the remaining EtOAc. A similar dissolution−concentration process was repeated twice, and the resulting solution was used for the following hydrolysis of methyl ester. An analytically pure sample of **10** was obtained by silica gel chromatography, followed by crystallization from MeCN. **10**: colorless needles, mp 185−205 °C; TLC $R_f$ 0.47 ($CH_2Cl_2$/EtOAc = 2/1); IR ($CHCl_3$) 2951, 1693, 1635, 1498, 1306 $cm^{-1}$; $^1H$ NMR ($CDCl_3$ + $CD_3OD$) $\delta$ 0.79 (s, 3), 0.83 (s, 3), 0.92 (s, 6), 0.95 (s, 3), 1.04 (s, 3), 1.0−2.0 (m, 22), 2.85−2.95 (m, 1), 3.83 (s, 3), 3.85−3.95 (m, 4), 4.19 (d, 1, $J$ = 13.5), 4.35 (d, 1, $J$ = 13.5), 5.59 (br s, 1), 6.31 (d, 1, $J$ = 15.8), 6.90 (dd, 1, $J$ = 8.7, 2.8), 6.93 (d, 1, $J$ = 15.4), 7.09 (d, 1, $J$ = 2.7), 7.23 (d, 1, $J$ = 15.4), 7.44 (d, 1, $J$ = 8.8), 7.75 (d, 1, $J$ = 15.8); $^{13}C$ NMR ($CD_3OD$) $\delta$ 15.2, 17.9, 18.7, 19.9, 22.7, 23.1, 23.2, 23.8, 24.3, 26.6, 30.8, 32.7, 32.8, 33.6, 33.9, 37.3, 37.3 40.3, 41.7, 42.3, 45.3, 45.8, 46.6, 49.0, 52.0, 53.7, 64.9, 65.0, 66.3, 112.9, 113.4, 118.5, 120.1, 127.4, 127.8, 129.0, 130.9, 132.2, 136.8, 138.0, 140.5, 157.2, 164.4, 166.3, 167.3, 180.8. $[\alpha]^{24}_D$ +62.8° ($c$ 1.006, $CHCl_3$). Anal. Calcd for $C_{46}H_{61}NO_{10}\cdot 1.2H_2O$: C, 68.24; H, 7.89; N, 1.73; $H_2O$, 6.36. Found: C, 67.98; H, 7.89; N, 2.02; $H_2O$, 6.39. HR-LSIMS $m/z$ 810.4195 $[M + Na]^+$ (calcd for $C_{46}H_{61}NO_{10}$-Na, 810.4190).

**Ketal Acid 11.** To a solution of **10** (4.62 mol) in dimethoxyethane (10 L) was added aqueous LiOH (10.6 kg in 9.6 kg of water) at −5 °C, and the resulting solution was stirred at the same temperature for 70 min. The mixture was poured into a mixture of water (38 L) and EtOAc (38 L), and the pH of the mixture was adjusted at 9.0 by adding 7% HCl (ca 2.9 kg). The separated organic layer was discarded, and EtOAc (38 L) was added to the aqueous layer. The pH of the mixture was acidified to 6.0 by adding 7% HCl with stirring, and the separated organic layer was washed with water (32 L) and then concentrated to 16 L. THF (48 L) was added to the residue, and the solution was again concentrated to 16 L. This process was repeated twice to give the THF solution of **11**, which was used for the next step. **11**: TLC $R_f$ 0.66 (EtOAc/AcOH/$H_2O$ = 60/1/1); $^1H$ NMR ($CD_3OD$) $\delta$ 0.81 (s, 3), 0.83 (s, 3), 0.90 (s, 3), 0.93 (s, 6), 0.96 (s, 3), 1.1−2.0 (m, 22), 2.85−2.95 (m, 1), 3.8−3.9 (m, 4), 4.16 (d, 1, $J$ = 12.5), 4.39 (d, 1, $J$ = 12.5), 5.58 (br s, 1), 6.36 (d, 1, $J$ = 16.0), 6.82 (d, 1, $J$ = 15.4), 6.89 (dd, 1, $J$ = 2.6, 8.7), 7.12 (d, 1, $J$ = 2.6), 7.18 (d, 1, $J$ = 8.7), 7.21 (d, 1, $J$ = 15.4), 7.67 (d, 1, $J$ = 16.0).

**Free Acid of S-0139 (7).** (+)-[27-[(*E*)-3-[2-[(*E*)-3-Carboxypropenoylamino]-5-hydroxyphenyl]propenoyloxy]-3-oxoolean-12-en-28-oic acid. To the THF solution of ketal acid **11** obtained as above was added 7% aqueous HCl (3.9 kg), and the resulting mixture was heated under reflux for 1 h. Water (38 L) and EtOAc (38 L) were added to the resulting solution, and the organic layer was separated, and then an aqueous layer was extracted with EtOAc (32 L). Organic layers were combined and extracted with 5% aqueous $NaHCO_3$ (27 kg ×3), and the aqueous extracts were combined. EtOAc (38 L) was added to the aqueous extracts, and the resulting two-layer solution was made acidic (pH < 4) by adding 7% HCl (26.5 kg). The two phases were separated, the aqueous layer was extracted with EtOAc (32 L), and the organic extracts were combined and washed with $H_2O$ (32 L ×2). To the resulting mixture was added EtOAc (48 L), and the solution was condensed under reduced pressure to 19 L. After a seed crystal of **7** was added and the solution stirred for 1 h, MeCN (16 L) was added at 70 °C, and the mixture was cooled to room temperature with stirring. The resulting thick slurry was filtered under a nitrogen atmosphere, and the collected precipitates were

washed with MeCN (6.4 L) and dried under reduced pressure to give **7** (2.43 kg, 72%). **7**: pale yellow needles (EtOAc), mp 207−215 °C; TLC $R_f$ 0.47 (EtOAc/AcOH/$H_2O$ = 60/1/1); HPLC (MeCN/$H_2O$/AcOH (50/50/0.1)) $t_R$ 7.2 min; IR (KBr) 3288, 2944, 1691, 1639, 1527, 1498, 1458 $cm^{-1}$; $^1H$ NMR ($CD_3OD$) $\delta$ 0.85 (s, 3), 0.87 (s, 3), 0.94 (s, 3), 1.03 (s, 6), 1.05 (s, 3), 1.05−2.05 (m, 20), 2.15−2.60 (m, 2), 2.85−3.00 (m, 1), 4.11 (d, 2, $J$ = 12.6), 4.49 (d, 2, $J$ = 12.6), 5.60 (s, 1), 6.36 (d, 2, $J$ = 16.0), 6.84 (d, 2, $J$ = 15.6), 6.89 (dd, 1, $J$ = 8.6, 2.6), 7.12 (d, 1, $J$ = 2.6), 7.14 (d, 1, $J$ = 8.6), 7.20 (d, 1, $J$ = 15.6), 7.66 (d, 1, $J$ = 16.0); $^{13}C$ NMR $\delta$ 15.8, 18.5, 20.7, 21.9, 23.9, 24.0, 24.5, 25.1, 27.2, 31.6, 33.5, 33.6, 33.9, 34.8, 34.9, 38.0, 40.2, 41.1, 42.6, 46.1, 46.8, 47.4, 48.4, 56.2, 66.6, 113.6, 119.4, 120.8, 128.0, 128.7, 129.8, 132.8, 132.9, 137.4, 138.7, 141.4, 158.0, 165.5, 168.0, 168.2, 181.5, 220.4. $[\alpha]^{24}_D$ +98.0° (*c* 1.002, $CH_3OH$). Anal. Calcd for $C_{43}H_{55}NO_9 \cdot 2.75H_2O$: C, 66.26; H, 7.82; N, 1.80; H2O, 6.36. Found: C, 66.37; H, 7.77; N, 1.88; $H_2O$, 6.39. HR-LSIMS *m/z* 730.3955 $[M + H]^+$ (calcd for $C_{43}H_{56}NO_9$, 730.3952).

**S-0139.** (+)-Disodium 27-[(*E*)-3-[2-[(*E*)-3-carboxypropenoylamino]-5-hydroxyphenyl]propenoyloxy]-3-oxoolean-12-en-28-oate. To a suspension of **7** (840.5 g, 1.08 mol) in $H_2O$ (2.9 L), aqueous 10% NaOH (927.5 g) was added dropwise with stirring over 1.5 h. The pH of the resulting solution was adjusted to 8.1 by adding aqueous 10% NaOH. The solution was filtered, and the resulting filtrate was freeze-dried to give S-0139 (837 g, 98%) as a pale yellow amorphous powder; TLC $R_f$ 0.50 (EtOAc/AcOH/$H_2O$ = 30/1/1); HPLC (PIC-A/MeCN (50/50)) $t_R$ 6.43 min; IR (KBr) 3420, 2863, 1690, 1633, 1570, 1497, 1466 $cm^{-1}$; $^1H$ NMR ($D_2O$, DSS standard) $\delta$ 0.72 (s, 3), 0.78 (s, 3), 0.86 (s, 3), 0.96 (s, 3), 0.98 (s, 6), 1.08−2.05 (m, 20), 2.15−2.60 (m, 2), 2.70−2.90 (m, 1), 4.01 (d, 2, $J$ = 12.7), 4.35 (d, 2, $J$ = 12.7), 5.56 (s, 1), 6.40 (d, 1, $J$ = 16.0), 6.88 (d, 2, $J$ = 15.4), 6.93 (d, 2, $J$ = 15.4), 7.01 (dd, 1, $J$ = 8.2, 2.6), 7.19 (d, 1, $J$ = 3.2), 7.20 (d, 1, $J$ = 8.4), 7.57 (d, 1, $J$ = 15.8); $^{13}C$ NMR ($D_2O$) $\delta$ 17.2, 20.3, 21.9, 23.6, 25.5, 25.9, 26.6, 26.7, 28.6, 32.7, 34.9, 34.9, 35.4, 35.4, 36.1, 39.1, 41.4, 42.0, 44.6, 47.9, 48.0, 50.2, 50.3, 50.7, 57.6, 68.9, 116.2, 122.4, 122.5, 128.5, 129.0, 132.2, 133.8, 134.8, 140.6, 141.0, 143.2, 160.5, 170.3, 171.3, 175.4, 189.2, 228.6. $[\alpha]^{25}_D$ +98.0° (*c* 1.001, $H_2O$). Anal. Calcd for $C_{43}H_{53}NO_9Na_2 \cdot 1.6H_2O$: C, 64.34; H, 7.06; N, 1.74; Na, 5.73; $H_2O$, 3.54. Found: C, 64.08; H, 7.18; N, 1.96; Na, 5.56; $H_2O$, 3.54.


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## Supporting Information Available

Copies of $^1H$ NMR spectra of compounds **6**, **11**, **12a**, **12b**, and **13** which lack elemental analyses (7 PDF). This material is available free of charge via the Internet at http://pubs.acs.org.


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