Agricultural and Biological Chemistry

ISSN: 0002-1369 (Print) (Online) Journal homepage: http://www.tandfonline.com/loi/tbbb19

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To cite this article: Akira Tanaka, Soichiro Otsuka & Kyohei Yamashita (1984) Biomimetic Total Synthesis of (–)-Aplysistatin, Agricultural and Biological Chemistry, 48:10, 2535-2540, DOI: 10.1080/00021369.1984.10866525

To link to this article: http://dx.doi.org/10.1080/00021369.1984.10866525

Agric. Biol. Chem., **48** (10), 2535~2540, 1984

Biomimetic Total Synthesis of (−)-Aplysistatin

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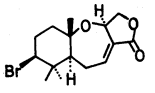
Received May 14, 1984

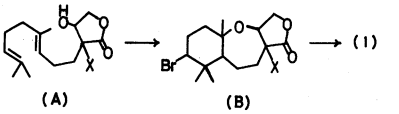
A biomimetic synthesis of (−)-aplysistatin (**1**) is described. The Wittig reaction of the keto ester **5** with homogeranyl triphenylphosphonium ylid gave the desired intermediate **3**. Successive treatment of **3** with activated manganese dioxide, sodium chlorite and aq. trifluoroacetic acid led to the unsaturated β-hydroxy lactone **2**, which was subjected to brominative cyclization to yield (−)-aplysistatin (**1**).

Aplysistatin (**1**), which was isolated from the sea hare *Aplysia angasi* in 1977,[1] is a bromine-containing sesquiterpenoid and shows significant inhibitory activity against murine lymphocytic leukemia P-388. Unique structural



(1)

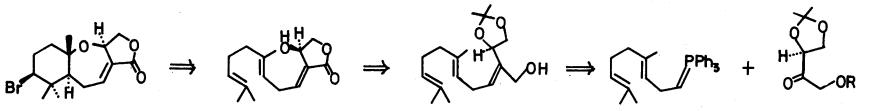
features, including an oxepane ring system, and the interesting biological activity of **1** have stimulated many synthetic chemists. Before our work, only one synthesis of racemic aplysistatin had been reported.[2a] Since then, three additional syntheses of **1** in racemic form[2b~d] and one chiral synthesis[2e] of **1** have been published. All of them have employed a variety of brominative cyclizations[3,4] involving concerted or stepwise processes to build up the bicyclic ether ring of the aplysistatin precursors, in which several further reaction steps toward the target molecule were required [*e.g.* (A)→(B)→ (**1**)]. We now wish to report a biomimetic synthesis directly leading to natural aplysistatin.



(A) (B)

Our synthetic strategy is outlined in Scheme I. Thus, condensation of the homogeranyl phosphonium ylid **4** with the chiral keto ester **5** (R = Ac or Bz) would provide the triene alcohol **3**, which would then be transformed into the β-hydroxy-α,β-unsaturated lactone **2**, a precursor of aplysistatin. Mercuric ion-initiated brominative cyclization of **2** can be expected to directly yield natural aplysistatin (**1**).

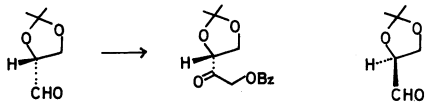
Our synthetic work began with the preparation of **5**. In our previous paper,[5] we described the synthesis of (*R*)-keto benzoate **7** from 1,2-*O*-isopropylidene-(*R*)-glyceraldehyde (**6**), which is easily accessible from D-mannitol.



(1) (2) (3) (4) (5)

SCHEME I.

Application of our approach to the preparation of the (*S*)-enantiomer **5** needed 1,2-*O*-isopropylidene (*S*)-glyceraldehyde (**8**)[6] or the corresponding (*R*)-alcohol[7] as a chiral progenitor. However, these are not as readily available as (*R*)-**6** and the corresponding (*S*)-alcohol. Consequently, we searched for other routes to **5**.

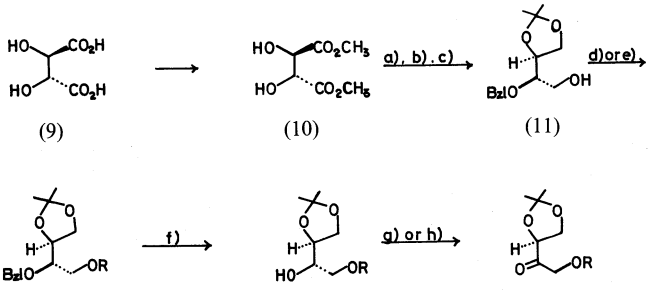


(6) (7) (8)

As depicted in Scheme II, we chose natural L-tartaric acid (**9**) as a logical chiral source of **5**, because **9** is inexpensive and possesses all of the carbon skeleton of **5**. Conversion of **9** into **11** was effected by following a recently reported procedure[6] involving slight modification to Ohno's synthesis.[8] The acetate **12a** and the benzoate **12b** were prepared from **11** by the conventional method. Then, **12a** was subjected to hydrogenolysis utilizing 10% Pd-C in ethanol at 50°C to yield the hydroxy acetate **13a** in a good yield. The Swern oxidation[9] of **13a** with oxalyl chloride and dimethylsulfoxide followed by a non-aqueous work-up[10] resulted in an excellent yield of the keto acetate **14a**. Attempted oxidation of **13a** with pyridinium dichromate[11] in dimethylformamide gave rise to only a low yield of **14a**, although a pure sample was obtained. Similarly, a smooth transformation of **11** into **14b** in the benzoate series was achieved. The (*S*)-keto benzoate **14b** thus obtained showed mp 58~59°C and $[\alpha]_D$ −80.2°, which agreed well with those of the (*R*)-enantiomer[5] except for the sign of rotation.

Construction of the diene moiety is shown in Scheme III. Since Hoye's procedure[12] to give **16** from **15** by using potassium cyanide in the presence of 18-crown-6 requires a long reaction time (6 days), we tried the substitution reaction under phase transfer conditions.[13] When allowed to react with aq. sodium cyanide and di-*n*-butylamine at room temperature, **15** provided an excellent yield of **16**. Transformation of **16** into homogeranyl bromide (**17**) was accomplished according to Cornforth's method.[14] The preparation of the requisite phosphonium salt **18** was effected by treating **17** with triphenylphosphine and suspended sodium bicarbonate in hot toluene, in which the lack of the bicarbonate resulted in the formation of cyclized products as proven by NMR. It may have been catalyzed by the hydrogen bromide that was probably generated by thermal decomposition of **17**.

With two requisite structural moieties in hand, we turned our attention to the convergent reaction steps toward aplysistatin (Scheme



(9) (10) (11)

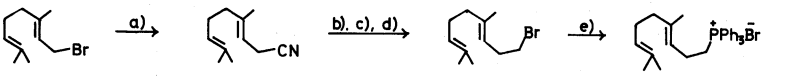
(12a)R = Ac
(12b)R = Bz

(13a)R = Ac
(13b)R = Bz

(14a)R = Ac
(14b)R = Bz

a) PhCHO, $HC(OCH_3)_3$, *p*-TsOH, PhH; b) $LiAlH_4$–$AlCl_3$, Et_2O–CH_2Cl_2; c) acetone, *p*-TsOH; d) Ac_2O, pyridine; e) PhCOCl, pyridine; f) H_2, 10% Pd–C, EtOH; g) $(COCl)_2$, DMSO, CH_2Cl_2; h) PDC, DMF.

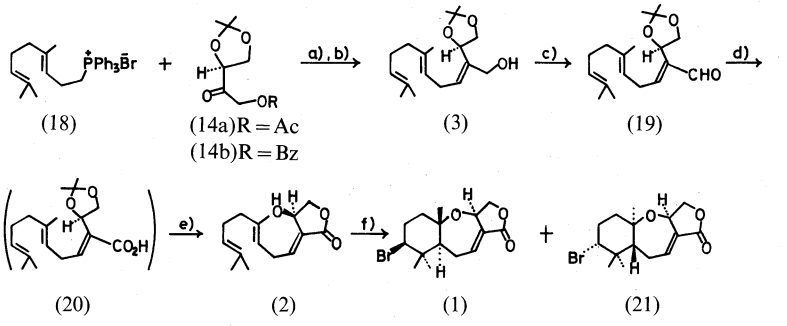
SCHEME II



(15) (16) (17)

a) aq. NaCN, di-*n*-butylamine; b) KOH, aq. EtOH; c) $LiAlH_4$, Et_2O; d) LiBr, acetone; e) PPh_3, $NaHCO_3$, toluene.

SCHEME III.



(18) (14a)R = Ac (14b)R = Bz (3) (19)

(20) (2) (1) (21)

a) *n*-BuLi, THF; b) KOH, CH_3OH; c) MnO_2, CH_2Cl_2; d) $NaClO_2$, 2-CH_3-2-butene, *t*-BuOH; e) 40% aq. CF_3CO_2H; f) $Hg(OCOCF_3)_2$, CH_3NO_2; KBr, H_2O; LiBr, Br_2, pyridine, O_2.

SCHEME IV.

IV). The Wittig condensation of 14a was carried out with the ylid derived from 18 and *n*-butyllithium in tetrahydrofuran at −10°C, and the resulting intermediate acetate was hydrolyzed without isolation. After chromatographic purification, the triene alcohol **3** was obtained in a 78% yield. The product thus obtained was found to be a mixture (*ca.* 6:4) of the geometrical isomers by HPLC, whereas **3** derived from the keto benzoate **14b** under similar reaction conditions consisted of *ca.* 8:2 of the isomeric mixture. No rigorous proof, however, for the expected geometry of the major isomer was available at this point (see below).

When subjected to oxidation with activated manganese dioxide at 0°C, **3** (isomer ratio 8:2) led to the aldehyde **19** in a 71% yield, along with recovery of a small amount of **3**. At a higher temperature, however, **3** resulted in the formation of a mixture of products containing significant quantities of by-products. In the NMR of **19**, the aldehydic proton signals were observed at 9.36 and 10.07 ppm in a ratio of 32:1. This means that the *Z*-isomer of **3** was exclusively oxidized to give the desired *E*-aldehyde **19** having high stereochemical purity and, consequently, the major alcohol obtained in the Wittig reaction possesses the requisite *Z* geometry.[15] Similarly, **3** (*Z*:*E*=6:4) gave, in a 50% yield, **19** having almost the same stereochemical purity as that mentioned above.

Oxidation of **19** to **20** presented some difficulties, because none of the generally used oxidants such as silver (I) oxide, silver (II) oxide, manganese dioxide/sodium cyanide and pyridinium dichromate gave satisfactory results. A solution to this problem was given by oxidation of **19** with sodium chlorite in aq. *t*-butyl alcohol in the presence of 2-methyl-2-butene.[16] The crude acid **20** obtained provided **2** (*E*:*Z*=*ca.* 13:1) in a 33% overall yield on exposure to aq. trifluoroacetic acid.

Finally, mercuric ion-induced brominative cyclization[2a] of **2** was carried out. Thus, upon successive treatment with mercuric trifluoroacetate in nitromethane and aq. potassium bromide, **2** gave rise to the bromomercury

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compound. This was then reacted with lithium bromide and bromine in pyridine to give (−)-aplysistatin, which was identical with a sample of (−)-**1** provided by Professor Pettit in all respects (mixed mp, optical rotation, IR, NMR and MS). Besides **1**, the minor isomer **21** was also isolated and was indistinguishable from (±)-12-epiaplysistatin on IR, NMR and MS spectra.

In addition to the chiral series described above, the synthesis of the racemic counterparts was also accomplished by employing (±)-keto benzoate (**14b**)[5] by way of the same sequence of reactions as that used in the chiral series. The melting point of racemic aplysistatin obtained in this way was not depressed on admixture with authentic (±)-**1** provided by Professor Hoye.

EXPERIMENTAL

All melting points and boiling points were uncorrected. Optical rotations were measured on JASCO DIP-4 spectrometer. IR spectra were taken on JASCO IRA-1 and IR-810 infrared spectrometers. ^{1}H-NMR spectra were measured on a JEOL JNM FX-100 Fourier transform spectrometer (99.5 MHz). MS spectra were recorded on a Hitachi M-52G spectrometer. HPLC was performed on a JASCO TRI ROTAR equipped with a UVIDEC-100-II detector.

(2S, 4′S)-2-Benzyloxy-2-(2′,2′-dimethyl-1′,3′-dioxolan-4′-yl)-1-ethanol (**11**). The compound was prepared from dimethyl L-tartrate following our recently reported procedure[6] involving a modification to Ohno's method.[8]

(2S, 4′S)-2-Benzyloxy-2-(2′,2′-dimethyl-1′,3′-dioxolan-4-yl)-ethyl acetate (**12a**). A solution of **11** (30.0 g) in pyridine (80 ml) was treated with Ac_2O (15.7 ml) in the usual manner. Distillation gave **12a** (33.3 g), bp 147°C (0.7 mmHg). $[\alpha]_D^{20}$ −12.4° (c=0.84, $CHCl_3$). IR ν_{max}^{film} cm^{-1}: 3050, 1750, 1500, 1390, 1380, 1240, 1080, 1050, 750, 700. NMR ($CDCl_3$) δ: 1.33 (3H, s), 1.40 (3H, s), 3.5∼4.0 (3H, m), 4.1∼4.3 (3H, m), 4.68 (2H, s), 7.30 (5H, s). *Anal.* Found: C, 65.14; H, 7.54. Calcd. for $C_{16}H_{22}O_5$: C, 65.29; H, 7.53%.

(2S, 4′S)-2-Benzyloxy-2-(2′,2′-dimethyl-1′,3′-dioxolan-4′-yl)-ethyl benzoate (**12b**). A solution of **11** (6.7 g) in pyridine (20 ml) was treated with benzoyl chloride (4 ml) in the usual way. The crude product obtained was passed through a short alumina column to afford the pure **12b** (9.1 g). $[\alpha]_D^{23}$ −9.3° (c=1.11, $CHCl_3$). IR ν_{max}^{film} cm^{-1}: 3080, 3040, 1730, 1600, 1590, 1500, 1385, 1375, 1275, 1120, 1100, 740, 720. NMR ($CDCl_3$) δ: 1.38 (3H, s), 1.45 (3H, s), 3.7∼4.1 (3H, m), 4.2∼4.5 (3H, m), 4.77 (2H, s), 7.30 (5H, m), 7.49 (3H, m), 7.97 (2H, m). MS m/z (rel. int.): 341 ($M^+ - CH_3$) (10), 298 (3), 255 (13), 192 (7), 176 (6), 149 (15), 133 (9), 105 (37), 101 (44), 91 (100), 43 (10).

(2S, 4′S)-2-(2′,2′-Dimethyl-1′,3′-dioxolan-4′-yl)-2-hydroxyethyl acetate (**13a**). Hydrogenolysis of **12a** (56.4 g) in 95% EtOH (300 ml) was effected by using 10% Pd-C (10 g) at 50°C. Distillation gave **13a** (34.6 g), bp 104°C (0.95 mmHg). $[\alpha]_D^{18}$ +2.4° (c=1.24, $CHCl_3$). IR ν_{max}^{film} cm^{-1}: 3460, 1750, 1390, 1380, 1240, 1075, 1060. NMR ($CDCl_3$) δ: 1.37 (3H, s), 1.45 (3H, s), 2.10 (3H, s), 2.53 (1H, d, J=6 Hz, OH), 3.7∼4.2 (8H, m). *Anal.* Found: C, 52.91; H, 7.99. Calcd. for $C_9H_{16}O_5$: C, 52.93; H, 7.90%.

(2S, 4′S)-2-(2′,2′-Dimethyl-1,′3′-dioxolan-4′-yl)-2-hydroxyethyl benzoate (**13b**). Hydrogenolysis of **12b** (15.7 g) in 95% EtOH (50 ml) was carried out using 10% Pd-C (3.1 g) at 50°C. The crude **13b** (11.4 g) was found to be almost pure on TLC. $[\alpha]_D^{23}$ +3.5° (c=1.02, $CHCl_3$). IR ν_{max}^{film} cm^{-1}: 3500, 3080, 1730, 1600, 1590, 1385, 1375, 1289, 1120, 1075, 720. NMR ($CDCl_3$) δ: 1.38 (3H, s), 1.46 (3H, s), 2.60 (1H, d, J=6.4 Hz, OH), 3.9∼4.4 (6H, m), 7.5 (3H, m), 8.0 (2H, m). MS m/z: 251 ($M^+ - CH_3$) (30), 165 (6), 131 (4), 105 (100), 101 (89), 77 (11), 43 (21).

(S)-2-(2′,2′-Dimethyl-1′,3′-dioxolan-4′-yl)-2-oxoethyl-acetate (**14a**).

a) *Pyridinium dichromate oxidation.* To a stirred mixture of pyridinium dichromate (4.2 g) in DMF (8 ml) was added a solution of **13a** (1.5 g) in DMF (2 ml) and stirring was continued for 48 hr at room temperature. After adding ether and powdered $MgSO_4$ followed by vigorous agitation of the mixture, the ether solution was decanted and the residue was thoroughly washed with ether. The combined solutions were washed with H_2O and dried. Evaporation gave a crude product which upon column chromatography (SiO_2) afforded **14a** (0.5 g). Short-path distillation yielded an analytical sample of **14a**. $[\alpha]_D^{16}$ −76.7° (c=1.82, $CHCl_3$) (lit.[17] $[\alpha]_D^{21}$ −64° (c=2.2, $CHCl_3$) for 88% *e.e.*). IR ν_{max}^{film} cm^{-1}: 1760, 1750, 1420, 1070. NMR ($CDCl_3$) δ: 1.39 (3H, s), 1.51 (3H, s), 2.16 (3H, s), 4.0∼4.3 (3H, m), 4.55 (1H, dd, J=6.0, 3.7 Hz), 4.95 (2H, ABq). *Anal.* Found: C, 53.54; H, 7.03. Calcd. for $C_9H_{14}O_5$: C, 53.46; H, 6.98%.

b) *Swern oxidation.*[9] To an activated DMSO reagent prepared from oxalyl chloride (3.7 ml) and DMSO (6.8 g) in $CHCl_2$ (155 ml) at between −60 and −50°C under an N_2 atmosphere was added dropwise a solution of **13a** (5.9 g) in CH_2Cl_2 (35 ml). After stirring for 1 hr, Et_3N (12.8 ml) was added slowly and the temperature was raised to 0°C. The mixture was diluted with ether and the resulting white ppt. was filtered. Evaporation gave a liquid which was passed through a short column of SiO_2.

Distillation afforded **14a** (5.0 g), bp 99°C (0.9 mmHg).

(S)-2-(2′, 2′-Dimethyl-1′, 3′-dioxolan-4′-yl)-2-oxoethyl-benzoate (**14b**). To an activated DMSO reagent prepared from oxalyl chloride (5.3 ml) and DMSO (9.7 g) in CH_2Cl_2 (230 ml) at between −60 and −50°C under N_2 was added dropwise a solution of **13b** (11.0 g) in CH_2Cl_2 (30 ml) and the mixture was stirred for one hour. Et_3N (18.2 ml) was added and the organic layer was separated. The aq. layer was extracted with CH_2Cl_2 and the combined solutions were washed sequentially with aq. $(CO_2H)_2$, aq. $NaHCO_3$ and sat. NaCl, and then dried. Evaporation afforded a crystalline solid which, upon recrystallization from ether/hexane, led to **14b** (6.0 g), mp 58∼59°C. $[\alpha]_D^{25}$ −80.2° ($c \times 1.18$, $CHCl_3$) (lit.[5]) for (*R*)-enantiomer, mp 56∼57°C, $[\alpha]_D^{25}$ +79.0° ($c=1.18$, $CHCl_3$)). *Anal.* Found: C, 63.35; H, 6.16. Calcd. for $C_{14}H_{16}O_5$: C, 63.62; H, 6.10%.

Geranyl cyanide (**16**). To a stirred solution of 33% aq. NaCN (160 ml) and di-*n*-butylamine (1.9 g) was added dropwise geranyl bromide[18] (**15**, 64.5 g) at 20∼25°C. After stirring for 2.5 hr, the aq. solution was extracted with ether in the usual manner. Distillation gave **16** (41.7 g), bp 90°C (4 mmHg) (lit.[12] bp 96°C (0.5 mmHg)). IR ν_{max}^{film} cm^{-1}: 2260, 1665, 830.

Homogeranyl bromide (**17**). This compound was prepared from **16** according to the literature procedure,[14] bp 85°C (2.5 mmHg) (lit. bp 64°C (0.4 mmHg)). IR ν_{max}^{film} cm^{-1}: 1660, 840.

Homogeranyltriphenylphosphonium bromide (**18**). A mixture of **17** (46.4 g) and Ph_3P (49.6 g) in toluene (110 ml) containing suspended $NaHCO_3$ (6.6 g) was refluxed for 6 hr. After adding hexane (110 ml), the whole mixture was shaken and the solution was decanted. The resulting highly viscous oil was repeatedly washed with benzene/hexane (1 : 1), dissolved in THF and filtered. Evaporation gave **18** (57.7 g), which was difficult to crystallize. Therefore, this was employed without further purification.

(S)-2-(2′, 2′-Dimethyl-1′, 3′-dioxolan-4′-yl)-6,10-dimethyl-2,5,9-undecatriene-1-ol (**3**).

a) *From the keto benzoate* **14b**. To an ylid solution prepared from **18** (14.2 g) and 1.5 M BuLi/hexane (17.4 ml) in THF (150 ml) was added dropwise a solution of **14b** (5.3 g) in THF (20 ml) at −10°C under N_2 and the reaction mixture was stirred overnight at room temperature. To this was added a solution of KOH (15 g) in CH_3OH (150 ml) and stirring was continued overnight. The total solution was poured into ice-cooled water and extracted with ether in the usual way. Column chromatography (SiO_2) of the crude product gave the desired **3** (4.6 g). HPLC (reversed phase SC-02 column, CH_3OH) showed two peaks in a ratio of *ca.* 8 : 2. IR ν_{max}^{film} cm^{-1}: 3440, 1660, 1385, 1375, 1060, 860. NMR ($CDCl_3$) δ: 1.41 (3H, s), 1.49 (3H, s), 1.61 (6H, s), 1.69 (3H, s), 2.01 (4H, broad s), 2.51 (1H, OH), 2.76 (2H, t, $J=7.6$ Hz), 3.65 (1H, t, $J=8.3$ Hz), 4.0∼4.3 (4H, m), 5.09 (2H, m), 5.63 (1H, t, $J=7.6$ Hz). *Anal.* Found: C, 73.15; H, 10.40. Calcd. for $C_{18}H_{30}O_3$: C, 73.43; H, 10.27%.

b) *From the keto acetate* **14a**. The Wittig reaction was carried out according to the procedure described in a). The product obtained in a 71% yield consisted of a mixture of isomers in a ratio of *ca.* 6 : 4 (HPLC: SC-02, CH_3OH).

(±)-**3** was also obtained from (±)-**14b** under conditions similar to those described above.

(S)-2-(2′, 2′-Dimethyl-1′, 3′-dioxolan-4′-yl)-6,10-dimethyl-2,5,9-undecatrien-1-al (**19**). To a well-stirred suspension of activated MnO_2 (36.0 g) in CH_2Cl_2 (70 ml) was added dropwise a solution of **3** (obtained from **14b**, 2.4 g) in CH_2Cl_2 (4 ml) at 0°C and stirring was continued at that temperature for 9 hr. Removal of MnO_2 by filtration and evaporation of the solvent gave a yellow oil which, upon chromatography on SiO_2, afforded **19** (1.7 g), together with the starting material **3** (0.6 g). The physical data of **19** are as follows. IR ν_{max}^{film} cm^{-1}: 2720, 1690, 1640, 1385, 1375, 1160, 1065, 860. NMR ($CDCl_3$) δ: 1.41 (3H, s), 1.51 (3H, s), 1.61 (3H, s), 1.68 (3H, s), 2.0 (4H, m), 2.68 (2H, broad), 3.36 (1H, t, $J=7.9$ Hz), 3.68 (1H, t, $J=7.8$ Hz), 4.25 (1H, t, $J=7.8$ Hz), 5.08 (2H, m), 6.56 (1H, t, $J=8$ Hz), 9.36 (32/33H, s), 10.07 (1/33H, s). *Anal.* Found: C, 73.78; H, 9.69. Calcd. for $C_{18}H_{28}O_3$: C, 73.93; H, 9.65%.

(±)-**19** was also prepared by a similar method.

(S)-2-(4,8-Dimethyl-3,7-nonadienylidene)-3-hydroxy-4-butanolide (**2**). To a solution of **19** (0.7 g) and 2-methyl-2-butene (5.1 ml) in *t*-BuOH (70 ml) was added dropwise a solution of $NaClO_2$ (1.1 g) and KH_2PO_4 (1.1 g) in H_2O (20 ml). After stirring at room temperature overnight, the mixture was treated with aq. $Na_2S_2O_3$. The volatile compounds were evaporated *in vacuo* and the aq. solution was exracted with ether in the usual manner after adjusting to pH 3 with dil. HCl. Evaporation gave the crude acid **20** (IR ν_{max}^{film} cm^{-1}: 3000∼2500, 1690, 1640, 1390, 1380, 1160, 1070, 860). This was then reacted with 40% aq. CF_3CO_2H (1 ml) at room temperature for 30 min to give a crude lactone which, upon column chromatography (SiO_2), afforded **2** (0.2 g). IR ν_{max}^{film} cm^{-1}: 3430, 1750, 1680, 1215, 995. NMR ($CDCl_3$) δ: 1.61 (3H, s), 1.68 (6H, s), 2.05 (4H, m), 2.56 (2H, m), 3.12 (1H, t, $J=7.6$ Hz), 3.57 (1H, broad OH), 4.17∼4.53 (2H, m), 5.08 (2H, 6.59 (1/14H, t, $J=7.6$ Hz), 6.92 (13/14H, t, $J=7.4$ Hz)). MS *m/z*: 250 (M^+) (2), 232 (1), 207 (4), 167 (12), 131 (19), 127 (27), 109 (12), 81 (56), 69 (100).

(±)-**2** was also obtained according to a similar procedure.

(−)-Aplysistatin (**1**) *and diasteromer* **21**. To a stirred solution of **2** (44 mg) in dry CH_3NO_2 (1 ml) was added

dropwise a solution of $Hg(OCOCF_3)_2$ (90 mg) in CH_3NO_2 at 0°C and stirring was maintained at room temperature. After 3 hr, the mixture was diluted with CH_2Cl_2 and the organic solution was washed with H_2O, dried and evaporated. The viscous residue was then treated with sat. aq. KBr solution (10 ml) in the dark overnight. The mixture was extracted with CH_2Cl_2 in the usual way to afford the bromomercury compound. This was dissolved in dry pyridine (2 ml) and the mixture was saturated with O_2. To this was added a solution of LiBr (30.6 mg) and Br_2 (0.01 ml) in pyridine (2 ml) pre-saturated with O_2 in the dark and the whole mixture was stirred overnight. After neutralizing with dil HCl, the aq. solution was extracted with ether in the usual way. The crude product was separated into (−)-aplysistatin (**1**, 4 mg) and its diastereomer **21** (2 mg) by preparative TLC. Repeated recrystallization from acetone/ hexane or toluene/hexane gave a sample of (−)-aplysistatin melting at 169～170°C that was identical in all respects with natural (−)-aplysistatin: mp 169～170°C (lit.[1] mp 173～175°C). $[\alpha]_D^{21}$ −355° (c=0.11, CH_3OH) (lit.[1] $[\alpha]_D^{25}$ −375° (CH_3OH); lit.[2e] $[\alpha]_D^{20}$ −421° (CH_3OH)). IR ν_{max}^{film} cm^{-1}: 3010, 2980, 2950, 2860, 1762, 1678, 1463, 1390, 1380, 1339, 1230, 1202, 1158, 1107, 1040, 1020, 1000, 990, 880, 743, 701, NMR ($CDCl_3$) δ: 0.96 (3H, s), 1.18 (3H, s), 1.29 (3H, s), 1.64 (2H, m), 1.8～2.3 (3H, m), 2.55 (2H, m), 3.95 (2H, m), 4.45 (1H, t, J=9 Hz), 5.51 (1H, m), 6.94 (1H, m). *MS m/z*: 330/328 (1.5/2), 300/298 (8/10), 249 (15), 231 (15), 291 (12), 139 (80), 123 (41), 121 (45), 91 (22), 83 (41), 69 (30), 43 (100). The diastereoisomer **21** was also indistinguishable from (±)-12-epiaplysistatin on IR, NMR and MS spectra. IR ν_{max}^{KBr} cm^{-1}: 1770, 1695, 1390, 1220, 1205, 1025, 1010. NMR ($CDCl_3$) δ: 0.94 (3H, s), 1.20 (3H, s), 1.42 (3H, s), 1.6～1.8 (3H, m), 2.11 (2H, m), 2.50 (2H, m), 3.93 (2H, m), 5.00 (1H, t, J=8.9 Hz), 5.18 (1H, m), 7.26 (1H, m). MS *m/z*: 330/328 (1/1), 315/313 (1/1), 300/198 (2/2), 249 (5), 231 (6), 219 (6), 204/202 (11/12), 139 (69), 123 (46), 95 (25), 83 (25), 81 (31), 69 (26), 43 (100).

(±)-Aplysistatin was also obtained following a method similar to that described above: mp 167°C, mixed mp 167°C.

Acknowledgments. We are grateful to Professor T. R. Hoye, University of Minnesota, for a sample and the spectra of (±)-aplysistatin and (±)-12-epiaplysistatin. We are also indebted to Professor G. R. Pettit, Arizona State University, for a sample of natural aplysistatin.

REFERENCES

1) G. R. Pettit, C. L. Herald, M. S. Allen, R. B. Von Dreele, L. D. Vanell, J. P. Y. Kao and W. Blake, *J. Am. Chem. Soc.*, **99**, 262 (1977).
2) a) T. R. Hoye and M. J. Kurth, *J. Am. Chem. Soc.*, **101**, 5065 (1979); b) J. D. White, T. Nishiguchi and R. W. Skeean, *J. Am. Chem. Soc.*, **104**, 3923 (1982); c) T. R. Hoye, A. J. Caruso, J. F. Dellaria, Jr. and M. J. Kurth, *J. Am. Chem. Soc.*, **104**, 6704 (1982); d) P. Gosselin et F. Rouessac, *Tetrahedron Lett.*, **24**, 5515 (1983); e) H.-M. Shieh and G. D. Prestwich, *Tetrahedron Lett.*, **23**, 4643 (1982).
3) For direct and indirect method of brominative cyclization: T. R. Hoye and M. J. Kurth, *J. Org. Chem.*, **44**, 3461 (1979) and references cited therein; A. Murai, A. Abiko, K. Kato and T. Masamune, *Chem. Lett.*, 1125 (1981); P. Gosselin et F. Rouessac, *Tetrahedron Lett.*, **23**, 5145 (1982); M. Riediker and J. Schwartz, *J. Am. Chem. Soc.*, **104**, 5842 (1982).
4) A recently developed mercury (II) triflate-amine complex leading to mercury-containing carbocycles in high yield and selectivity is of potential utility because of easy replacement of mercury to other functional groups including bromide. See M. Nishizawa, H. Nishide and Y. Hayashi, *Tetrahedron Lett.*, **24**, 2581 (1983).
5) A. Tanaka and K. Yamashita, *Agric. Biol. Chem.*, **44**, 199 (1980). The melting points recorded for (+)- and (±)-keto benzoate in this report should be 56～57°C and 71～72°C, respectively.
6) A. Tanaka, S. Otsuka and K. Yamashita, *Agric. Biol. Chem.*, **48**, 2135 (1984).
7) For example, S. Takano, H. Numata and K. Ogasawara, *Heterocycles*, **19**, 327 (1982) and references cited therein.
8) K. Fujita, H. Nakai, S. Kobayashi, K. Inoue, S. Nojima and M. Ohno, *Tetrahedron Lett.*, **23**, 3507 (1982).
9) A. J. Mancuso and D. Swern, *Synthesis*, 165 (1981).
10) The aqueous workup generally used in the Swern oxidation decreased the yield of **14a** to a considerable extent. See, ref. 9.
11) E. J. Corey and G. Schmidt, *Tetrahedron Lett.*, 399 (1979).
12) T. R. Hoye and M. J. Kurth, *J. Org. Chem.*, **43**, 3693 (1978).
13) W. P. Reeves and M. R. White, *Synth. Commun.*, **6**, 193 (1976).
14) J. W. Cornforth, R. H. Cornforth and K. K. Mathew, *J. Chem. Soc.*, 2539 (1959).
15) K. C. Chan, R. A. Jewell, W.H. Nutting and H. Rapoport, *J. Org. Chem.*, **33**, 3382 (1968).
16) B. S. Bal, W. E. Childers, Jr. and H. W. Pinnick, *Tetrahedron*, **37**, 2091 (1981).
17) T. Shono, H. Ohmizu and N. Kise, *Tetrahedron Lett.*, **23**, 4801 (1982).
18) P. A. Grieco and Y. Masaki, *J. Org. Chem.*, **39**, 2135 (1974).