Synthesis of Both the Enantiomers of Phoracantholide I, A Defensive Secretion of the Eucarypt Longicorn (*Phoracantha synonyma*), Employing Microbial Asymmetric Reduction with Immobilized Baker's Yeast[1)]

Yoshinobu NAOSHIMA,* Hidenobu HASEGAWA,† Tadashi NISHIYAMA, and Akihiro NAKAMURA
Department of Biological Chemistry, Faculty of Science, Okayama University of Science, 1-1 Ridai-cho, Okayama 700
†Midori Kagaku Co., Ltd., 19-12 Kamiochiai, Shinjuku-ku, Tokyo 161
(Received July 19, 1988)

Synopsis. Highly optically pure (*R*)- and (*S*)-phoracantholide I were synthesized in relatively short steps starting from diethyl 3-oxoglutarate by means of a microbial asymmetric reduction of the intermediate keto acid with immobilized baker's yeast entrapped in gels of κ-carrageenan.

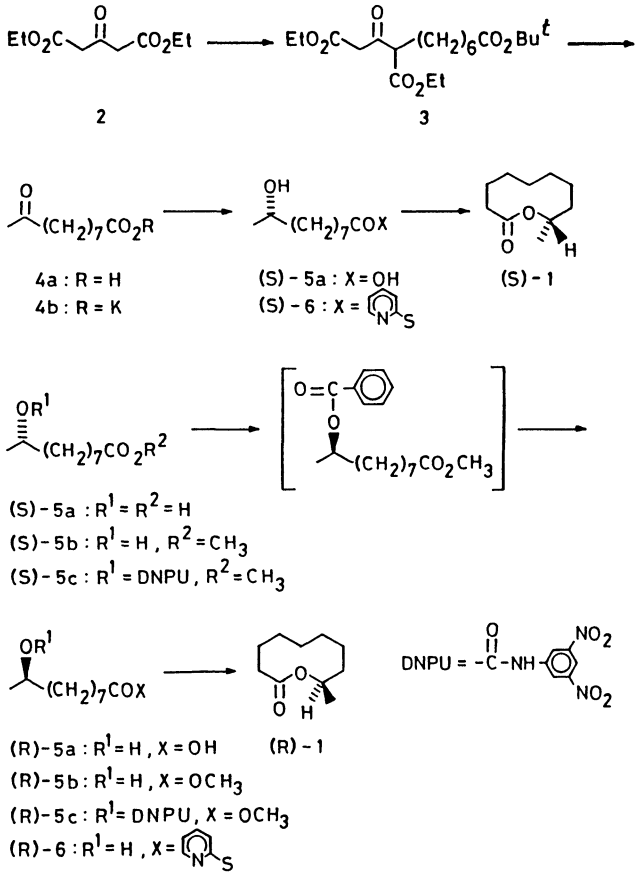
Since phoracantholide I (**1**), possessing a ten-membered lactone ring, has been isolated as a defensive secretion from the metasternal gland of the eucarypt longicorn, *Phoracantha synonyma*,[2)] several synthetic methods were reported for racemic **1**.[3)] The first synthesis of optically active **1** has recently been achieved by Kitahara et al. and the (*R*)-enantiomer was announced as the natural form of a defensive secretion of the insect.[4)] However, the method required multiple steps and the optical purity of synthesized enantiomers based on their GLC analyses with a chiral stationary phase was 89% e.e.

Recently we synthesized each enantiomer of 5-hexadecanolide and 4-dodecanolide with high optical purities via asymmetric reduction with immobilized baker's yeast (IBY).[5)] These are the pheromone of the oriental hornet and a defensive secretion of rove beetles. IBY, compared with free baker's yeast, has been found to have advantages in terms of the separation of products from the catalyst and of the reuse of the catalyst.[5,6)] In the expansion of the horizons of IBY in organic synthesis, we report herein a facile and short-step synthesis of the two enantiomers of **1** with highly optically pure forms.

Results and Discussion

First, diethyl 3-oxoglutarate (**2**) was alkylated regioselectively with *t*-butyl 7-bromoheptanoate and $Mg(OEt)_2$ to give the monoalkylated 3-oxoglutarate **3**, which was converted by a decarboxylative hydrolysis to the keto acid **4a** with a 78% yield from **2**. Compound **4a**, after treatment with an aqueous solution of KOH, was subjected to a microbial asymmetric reduction in an aqueous solution of KCl with the IBY entrapped by κ-carrageenan[5)] to give an optically pure alcohol (*S*)-**5a**. The optical purity of (*S*)-**5a** was determined to be 96% e.e. by HPLC analysis of the corresponding 3,5-dinitrophenylurethane (DNPU) derivative (*S*)-**5c**. Finally, (*S*)-**5a** was lactonized via the corresponding pyridinethiol ester (*S*)-**6** in the presence of $AgClO_4$ to give (*S*)-**1** with a 44% yield. For the synthesis of (*R*)-**1** reported as the natural form of **1**, the methyl ester (*S*)-**5b** was converted via a Mitsunobu inversion to (*R*)-**5a** with about 95% e.e. This was based on the HPLC analysis of a DNPU derivative (*R*)-**5c**. Lactonization of (*R*)-**5a**, as described above, gave the desired (*R*)-**1**. Thus highly optically pure (*R*)- and (*S*)-**1** were synthesized in seven and four steps, respectively, starting from **2**. The optical purity of the two enantiomers should be at least 95% e.e., because it was assumed that no apparent racemization at the chiral center of (*R*)- and (*S*)-**5a** occurred during their lactonization.

As shown in Table 1 the reuse of the IBY was achieved in two solvent systems, one of which is an aqueous KCl solution and the other an aqueous KCl solution containing ethyl acetate (EA). The reproducibility of both the chemical and optical yields was examined in each system. For the aqueous KCl solution system the first use of IBY catalyst gave a low chemical yield of (*S*)-**5a**, but after the 3rd use the yield was raised to 41%, a maximum yield of 46% being obtained after the 4th use. It is of interest that the



Scheme 1.

Table 1. Reuse of Immobilized Baker's Yeast in the Asymmetric Reduction of 9-Oxodecanoic Acid (**4a**)

Recycle number	Yield of **5a** (%)		Optical purity of (*S*)-**5a** (% e.e.)	
	AK[a]	AKEA[b]	AK[a]	AKEA[b]
1	23	22	96	96
2	28	26	96	95
3	41	31	96	96
4	46	38	96	96
5	32	21	96	94
6	30	—	96	—
7	15	—	96	—

a) AK: 2% aqueous KCl solution. b) AKEA: 2% aqueous KCl solution containing ethyl acetate.

optical purity of the product (*S*)-**5a** remained almost constant, 96% e.e., throughout all the repetitive use of the catalyst. An EA-containing aqueous solution system with a carboxylic acid **4a** as the substrate also produced (*S*)-**5a**, and the optical purity was about the same as that observed for the aqueous solution system employing a potassium salt **4b**. Further, the trend of the chemical yield for EA-containing aqueous solution system was comparable with that obtained for aqueous solution system, the yield of **5a** attaining a maximum of 38% after the 4 th use.

It is noteworthy that (1) the IBY catalyst entrapped with κ-carrageenan can be stored in a 2% aqueous solution of KCl for six or more months at 0—5 °C, (2) the catalyst is reusable many times during the period, (3) the optical purity of the product is reproducible throughout all of the use of the catalyst, (4) the catalyst can be recovered and separated easily from the reaction mixture by simple filtration.

Experimental

General. IR spectra were determined on a Hitachi 260-10 spectrometer and on a Perkin Elmer 1720 Fourier transform IR spectrometer. ^{1}H and ^{13}C NMR spectra were obtained on a Hitachi R-24B spectrometer and on a Hitachi R-90H Fourier transform NMR spectrometer, respectively, in $CDCl_3$ solutions with tetramethylsilane as an internal standard. EI and CI mass spectra were recorded on a JEOL JMS-D300 mass spectrometer at 70 eV and 200 eV (isobutane), respectively, using a direct insertion probe. Optical rotations were measured on a Horiba SEPA-200 high sensitivity polarimeter. Column chromatography was carried out with 70—230 mesh silica gel (Merck Kieselgel 60 Art 7734).

Determination of the Optical Purities of (*R*)- and (*S*)-5a. (*R*)- and (*S*)-**5a** were converted via their methyl esters to (*R*)- and (*S*)-**5c** by treating with 3,5-dinitrophenyl isocyanate. HPLC analyses were carried out on a Gasukuro Kogyo model 576 liquid chromatograph equipped with a UV detector (254 nm). A Sumipax 2100 4×250 mm column (Sumitomo Chemical Co., Ltd., Osaka) was used with hexane: 1,2-dichloroethane:ethanol=100:20:1 (v/v, flow rate, 1 mL min^{-1}) as solvent. (*R*)- and (*S*)-**5c** were separable under these conditions and each showed two peaks. (*R*)-**5c**: R_t 12.4 min (2.5%) and 13.7 min (97.5%). (*S*)-**5c**: R_t 12.4 min (98%) and 13.7 min (2%). The optical purities of (*R*)- and (*S*)-**5a** were therefore 95% e.e. and 96% e.e.

9-*t*-Butyl Diethyl 2-Oxo-1,3,9-nonanetricarboxylate (3). Compound **3** was synthesized by alkylating diethyl 3-oxoglutarate (**2**, 20.2 g, 100 mmol) with *t*-butyl 7-bromoheptanoate (29.2 g, 110 mmol) in the presence of $Mg(OEt)_2$ according to the procedures described previously.[7] Purification by column chromatography on silica gel (hexane:ethyl acetate=3:1, v/v) gave **3** as a pale yellow viscous liquid (33.9 g, 88%). IR (neat) 1730, 1715 cm^{-1}. ^{1}H NMR δ=1.12—1.50 (21H, s at 1.43), 3.43 (2H, s), 3.55 (1H, t, *J*=7.2 Hz), 4.15 (4H, q, *J*=7 Hz). ^{13}C NMR δ=197.33, 172.82, 169.01, 166.44, 79.79. EIMS *m/z* (%) 330 (M^+ −56, 1), 313 (M^+ −73, 13), 202 (41), 156 (73), 57 (100). CIMS *m/z* (%) 387 [$(M+H)^+$, 41], 331 [$(M+H-56)^+$, 100].

9-Oxodecanoic Acid (4a). Monoalkylated 3-oxoglutarate **3** (19.3 g, 50 mmol) was heated in a 15% aqueous solution of NaOH (360 mL) under reflux for 15 h. The hydrolyzed mixture was acidified with concd HCl, saturated with NaCl and extracted with ether. Work-up of the ethereal solution gave a crude solid, which was recrystallized from petroleum ether to give **4a** as needles (8.28 g, 89%), mp 41—42 °C. IR (KBr) 3100, 1700 cm^{-1}. ^{1}H NMR δ=1.14—1.88 (10H, m), 2.15 (3H, s), 8.96 (1H, s). ^{13}C NMR δ=208.89, 179.09, 43.57. EIMS *m/z* (%) 186 (M^+, 2), 168 (M^+ −18, 6), 111 (20), 58 (70), 43 (100). CIMS *m/z* (%) 187 [$(M+H)^+$, 100]. Found: C, 64.38; H, 9.89%. Calcd for $C_{10}H_{18}O_3$: C, 64.69; H, 9.74%.

(*S*)-(+)-9-Hydroxydecanoic Acid [(*S*)-5a]. Use of Aqueous Solution System: According to the procedures described previously,[1,5] a mixture of the potassium salt **4b** derived from **4a** (1 g), baker's yeast (50 g) immobilized in carrageenan beads, and D-glucose (40 g) in a 2% aqueous solution of KCl (1000 mL) was shaken at 30 °C. Work-up of the reaction mixture gave crude (*S*)-**5a**, which was purified by column chromatography on silica gel (hexane:ethyl acetate=1:1, v/v) to give (*S*)-**5a** as a colorless liquid (465 mg, 46%), $[\alpha]_D^{22}$ +8.14° (*c* 0.41, chloroform). IR (neat) 3500, 1710 cm^{-1}. ^{1}H NMR δ=1.18 (3H, d, *J*=6 Hz), 1.12—1.86 (12H, m), 2.25 (4H, m), 3.72 (1H, m), 7.08 (2H, s). ^{13}C NMR δ=178.85, 68.08. EIMS *m/z* (%) 170 (M^+ −18, 1), 115 (17), 101 (41), 45 (100). CIMS *m/z* (%) 189 [$(M+H)^+$, 24], 171 [$(M+H-18)^+$, 100]. Found: C, 63.72; H, 10.91%. Calcd for $C_{10}H_{20}O_3$: C, 63.79; H, 10.71%.

Use of EA-Containing Aqueous Solution System: The same procedure was followed as described above except for using a 2% aqueous solution of KCl (1000 mL) containing ethyl acetate (10 mL) as solvent and the keto acid **4a** (1 g) itself as substrate.

(*S*)-(+)-Phoracantholide I [(*S*)-1]. To a solution of (*S*)-**5a** (160 mg, 0.85 mmol) in dry benzene (1 mL) were added triphenylphosphine (320 mg, 1.22 mmol) and dipyridyl disulfide (270 mg 1.22 mmol) under argon and the mixture was stirred for one hour at room temperature. The resulting pyridinethiol ester (*S*)-**6** was dissolved in dry benzene (50 mL) and the solution was added dropwise to a stirred suspension of $AgClO_4$ (1.13 g) in refluxing benzene (120 mL) under argon over period of 8 h with a motor-drive syringe (Micro Feeder JP-V, Furue Science, Tokyo). After the addition had been completed, the mixture was refluxed for an additional 15 min and cooled to room temperature. The precipitate was filtered off and washed with ether. The filtrate and washings were combined and concentrated in vacuo. The residue was purified by column chromatography on silica gel (hexane: ethyl acetate=10:1, v/v) followed by fractional distillation to give (*S*)-**1** (63 mg, 44%) with a characteristic odor, bp 65 °C (bath temperature)/3 mmHg (1 mmHg=133.32 Pa), $[\alpha]_D^{22}$ +38.38° (*c* 0.06, chloroform) [lit,[4] $[\alpha]_D^{22}$ +38.8° (*c* 0.68, chloroform)]. IR (neat) 1725 cm^{-1}. ^{1}H NMR δ=1.25 (3H, d, *J*=6 Hz), 4.72—5.51 (1H, m). EIMS *m/z* (%) 170 (M^+, 3), 152 (M^+ −18, 17), 126 (12), 111 (11), 98 (100), 84 (50), 69 (30), 54 (64), 40 (55). CIMS *m/z* (%) 171 [$(M+H)^+$, 100]. Found: C, 70.32; H, 10.63%. Calcd for $C_{10}H_{18}O_2$: C, 70.54; H, 10.66%.

(*R*)-(−)-9-Hydroxydecanoic Acid [(*R*)-5a]. Methyl ester (*S*)-**5b** (220 mg, 1.1 mmol) prepared from (*S*)-**5a** was dissolved in dry THF (15 mL) and the solution was stirred under ni-

trogen while triphenylphosphine (600 mg, 2.2 mmol) and benzoic acid (300 mg, 1.6 mmol) were added. To the stirred solution was slowly added diethyl azodicarboxylate (400 mg, 2.2 mmol) and the mixture was allowed to stir at room temperature overnight. After evaporation of the solvent in vacuo, the residue was chromatographed on silica-gel column. Elution with benzene gave a benzoate ester, a solution of which in THF (2 mL) was added dropwise over a 10-min period to a stirred and ice-cooled 1 mol dm^{-3} aqueous solution of KOH (2 mL) containing methanol (1.5 mL). After being stirred for 30 min at room temperature, the mixture was acidified with concd HCl to pH 1—2 under ice-cooling and extracted with dichloromethane. Work-up of the organic layer gave a crude product, which was purified by column chromatography on silica gel (hexane : ethyl acetate=2 : 1, v/v) followed by preparative TLC (hexane : ethyl acetate=1 : 1, v/v) to give (*R*)-**5a** as a colorless liquid (112 mg, 54% from (*S*)-**5a**), $[\alpha]_D^{22}$ −8.13° (*c* 0.11, chloroform). The IR, NMR, and CI mass spectra were identical with those of (*S*)-**5a**. Found: C, 63.66; H, 10.78%. Calcd for $C_{10}H_{20}O_3$: C, 63.79; H, 10.71%.

(*R*)-(−)-Phoracantholide I [(*R*)-1]. Lactonization of (*R*)-**5a** (94 mg, 0.5 mmol), as described for (*S*)-**5a**, gave (*R*)-**1** (28 mg, 33%), $[\alpha]_D^{22}$ −37.4° (*c* 0.02, chloroform) [lit,[4] $[\alpha]_D^{22}$ −35.1° (*c* 1.15, chloroform)]. (*R*)-**1** was identical in its spectral data with (*S*)-**1**. Found: C, 70.29; H, 10.81%. Calcd for $C_{10}H_{18}O_2$: C, 70.54; H, 10.66%.

We wish to thank Dr. Seiichiro Kawabe, Department of Fundamental Natural Science, Okayama University of Science, for his advice concerning the immobilization of baker's yeast. The present work was partially supported by the Agricultural Chemical Research Foundation and by a Grant-in-Aid for Scientific Research No. 63560135 from the Ministry of Education, Science and Culture.

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