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An Improved Synthesis of the New Angiotensin Converting Enzyme Inhibitor CV-5975 *via* a Chemoenzymatic Process

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A chemoenzymatic synthesis of the new angiotensin converting enzyme inhibitor CV-5975 (**1**) is described. The optically active key intermediate for the synthesis of **1**, ethyl (*R*)-6-(1-benzyloxycarbonyl-4-piperidyl)-2-hydroxyhexanoate ((*R*)-**4**), was prepared by kinetic resolution of the racemic α-hydroxyester ((*RS*)-**4**) with a lipase and also by asymmetric reduction of the α-oxoester (**3**) with baker's yeast. The enantiomeric excess (ee) of the α-hydroxyester ((*R*)-**4**) produced by these enzymatic procedures exceeded 60%. This optically active alcohol ((*R*)-**4**) was converted to its mesylate ((*R*)-**5**), which was then subjected to S_N2 reaction with the aminobenzothiazepine derivative (**2**) followed by deprotection to yield **1**.

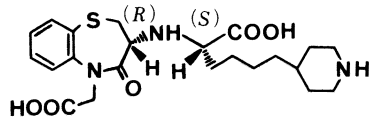
Keywords——ACE inhibitor; 1,5-benzothiazepine derivative; chemoenzymatic synthesis; enzymatic hydrolysis; baker's yeast reduction; CV-5975

Recently, we reported the discovery of the new angiotensin converting enzyme (ACE) inhibitor, (*R*)-3-[(*S*)-1-carboxy-5-(4-piperidyl)pentyl]amino-4-oxo-2,3,4,5-tetrahydro-1,5-benzothiazepine-5-acetic acid (CV-5975; **1**), which shows potent and long-lasting *in vivo* inhibitory activity.[1a–c] This compound includes two asymmetric carbons, with (*R*)-configuration of the 3-position and (*S*)-configuration in the side chain, both of which have been confirmed to be essential for the high biological activity.[1c] Introduction of (*R*)-chirality into the 3-position of the benzothiazepine skeleton, leading to the optically pure intermediate **2** (ee, >99%),[2] was achieved by using a natural amino acid, L-cysteine, as the starting material as described in our earlier report.[1a] However, little asymmetric induction was observed in the reductive alkylation of **2** with the α-oxoester **3** in the presence of sodium cyanoborohydride ($NaBH_3CN$), producing a diastereomeric mixture of (*R*),(*S*)-**6** and (*R*),(*R*)-**6** (*ca.* 1 : 1) in 19% yield.[1c]

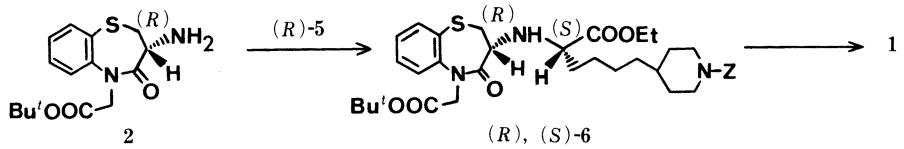
A more efficient and practical route to **1** was needed in order to make the compound more available for further biological studies. In our previous work,[1c] we found that a substitution reaction of the α-methanesulfonyloxyester (*RS*)-**5** with **2** proceeded smoothly to yield a diastereomeric mixture of diesters ((*R*),(*S*)-**6** and (*R*),(*R*)-**6**) in good yield. This suggested that if the (*R*)-α-methanesulfonyloxyester (*R*)-**5** could be obtained, it would react with **2** in an S_N2 process without racemization, thus giving the desired diester (*R*),(*S*)-**6** exclusively.[3,4]

This report describes an improved synthesis of CV-5975 (**1**) which utilizes chemoenzymatic methods to prepare the key chiral fragment, ethyl (*R*)-6-(1-benzyloxycarbonyl-4-piperidyl)-2-hydroxyhexanoate ((*R*)-**4**).

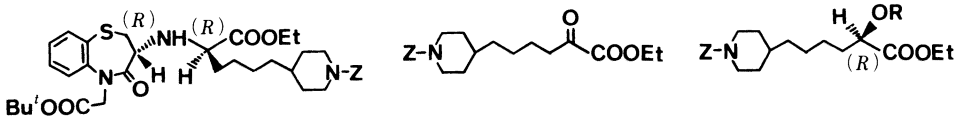
Initially, we attempted to resolve the racemic α-hydroxyacid (*RS*)-**7** by salt formation with a variety of chiral amines[5] available commercially. Although quinine and cinchonidine formed crystalline salts of the acid, complete resolution could not be attained by recrystallization because there were only small differences in the relative solubilities. This



1 : CV-5975



2 (*R*)-5 (*R*), (*S*)-6 1



(*R*), (*R*)-6 3 (*R*)-4 : R=H
(*R*)-5 : R=Ms

(*RS*)-4 : R_1=H, R_2=Et
(*RS*)-5 : R_1=Ms, R_2=Et
(*RS*)-7 : R_1=H, R_2=H

8

Bu^t=*tert*-Bu

Z=$COCH_2Ph$

Chart 1

unsuccessful approach prompted us to examine kinetic resolution[6] of the racemic α-hydroxyester (*RS*)-**4** utilizing hydrolytic enzymes to produce the chiral α-hydroxyester (*R*)-**4**. First, we screened a variety of hydrolytic enzymes[7] for activity towards the substrate (*RS*)-**4** and found that lipase M-AP10 (from *Mucor*)[8] and lipase PN (from *Phycomyces nitens*)[9] could hydrolyze the ester moiety of (*RS*)-**4** without any side reactions. Next, the relation between the conversion (%) and the optical purity (% ee) of the remaining substrate fraction was investigated with lipase M-AP10 and lipase PN. The hydrolysis was carried out with vigorous mixing of the substrate (*RS*)-**4** and the enzyme in 0.1 M phosphate buffer (pH 6.8) at 30 °C.[10] At suitable intervals, the conversion (%) was monitored by analyzing an aliquot of the reaction mixture by high-performance liquid chromatography (HPLC). The enantiomeric excess (% ee) of the unhydrolyzed ester in the sample was also determined by HPLC after formation of the corresponding (*S*)-(−)-α-methoxy-α-(trifluoromethyl)phenylacetic acid

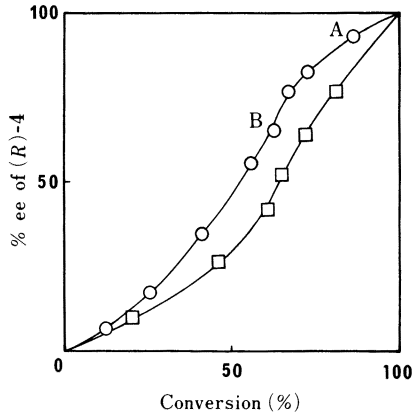


Fig. 1. The Relation between Conversion (%) and % ee of the Remaining α-Hydroxyester ((*R*)-**4**)

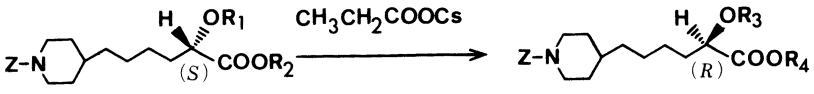
—○—, lipase PN; —□—, lipase M-AP10.

(MTPA) ester **8**.[11] As shown in Fig. 1,[12] lipase PN showed a slightly higher stereoselectivity than lipase M-AP10. The absolute configuration of the remaining unreacted α-hydroxyester was assigned as *R* as follows: highly optically active ester (93% ee) was obtained by stopping the hydrolysis when the conversion reached 86% (lipase PN, point A in Fig. 1). This ester was converted to the mesylate **5** which was allowed to react with **2** (90 °C, 20 h) to yield the diester **6**, which proved to be identical with (*R*),(*S*)-**6** on HPLC. Furthermore, this product showed high diastereomeric excess (de, 93%), which corresponded to 93% ee of the α-hydroxyester **4**. These findings indicated that complete Walden inversion had occurred at the α-position of the mesylate in this S_N2 process and therefore, the configuration of the α-hydroxyester should be *R*.

To use the resolved ester in the practical synthesis of **1**, we scaled up the procedure to a run with 40 g of the substrate (*RS*)-**4**. The reaction was stopped at 63% conversion (point B, in Fig. 1) to obtain (*R*)-**4** in a suitable chemical yield (14.6 g, 37%). The use of this (*R*)-enriched α-hydroxyester (64% ee) allowed us to omit chromatographic separation of the diastereomers **6** in the course of synthesis of CV-5975 (**1**). Treatment of this (*R*)-**4** with mesyl chloride (pyridine, 0 °C) afforded the mesylate (*R*)-**5**, which was allowed to react with **2** to yield the diester in 85% yield with 80:20 ((*R*),(*S*)-**6**: (*R*),(*R*)-**6**) diastereomeric ratio. Deprotection of this diester (HBr–AcOH; aqueous NaOH) gave CV-5975 (**1**), which was readily purified by recrystallization from water.

The above enzyme system hydrolyzed the (*S*)-ester to produce the (*S*)-acid ((*S*)-**7**), leaving the exploitable (*R*)-enriched ester ((*S*)-**4**) behind. Thus, methods are also needed to utilize the (*S*)-acid ((*S*)-**7**) for the synthesis of **1**. This was achieved by Swern oxidation[13] of its ester ((*S*)-**4**) into the α-oxoester **3**, which could be reduced to obtain the starting substrate (*RS*)-**4**.[1c] Also, hydroxy group inversion of the (*S*)-ester ((*S*)-**4**, 74% ee) could be accomplished *via* Kellogg's method[14] ((*S*)-**7**→(*S*)-**4**→(*S*)-**5**→(*R*)-**9**→(*R*)-**7**→(*R*)-**4**) and the Mitsunobu reaction[15] ((*S*)-**7**→(*S*)-**4**→(*R*)-**10**) as shown in Chart 2. These inversions proceeded without perceptible racemization as shown by comparison of ee on HPLC or the specific rotation of the materials (*S*-configuration) with that of the products (*R*-configuration).

Our next approach to preparing the (*R*)-hydroxyester (*R*)-**4** was asymmetric reduction of the α-oxoester **3**. One of the most promising methods for the preparation of an optically active alcohol seems to be baker's yeast reduction of a carbonyl compound.[16] This method is very versatile for organic synthesis because it is simple and economical as well as highly enantioselective. The stereochemistry of the alcohol produced by baker's yeast reduction can be predicted by the so-called Prelog rule[17] in the case of β-oxocarboxylic acid derivatives.[16]



(*S*)-**7** : $R_1=R_2=H$
(*S*)-**4** : $R_1=H$, $R_2=Et$
(*S*)-**5** : $R_1=Ms$, $R_2=Et$

(*R*)-**9** : $R_3=COCH_2CH_3$, $R_4=Et$
(*R*)-**7** : $R_3=R_4=H$
(*R*)-**4** : $R_3=H$, $R_4=Et$

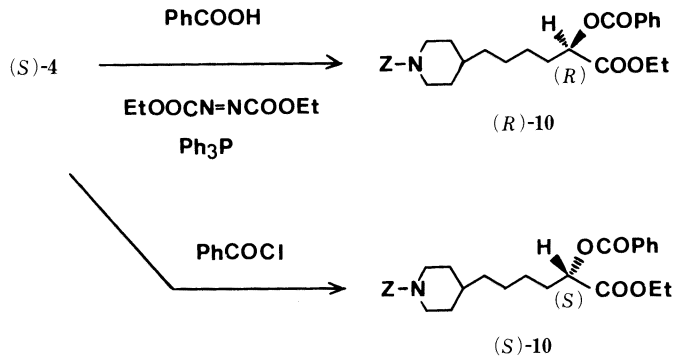
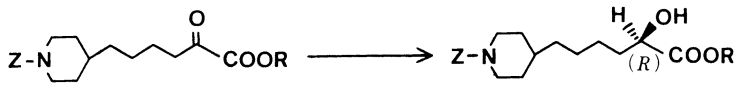


Chart 2

TABLE I. Baker's Yeast Reduction of α-Oxoesters (**3**, **11a**—**c**)



3, **11a**—**c** (*R*)-**4**, **12a**—**c**

Entry	α-Oxoester R	(g)	Reaction time (h)	(*R*)-α-Hydroxyester yield (%)[a]	((*R*)-**4**, **12a**—**c**) % ee
1	Me (**11a**)	(1)	22	52	37
2	Et (**3**)	(1)	46	34	54
3	Pr (**11b**)	(1)	42	43	32
4	Bu (**11c**)	(1)	66.5	31	30
5	Et (**3**)	(40)	48	46	61

a) Isolated yield.

However, few studies have been done with α-oxocarboxylic acid derivatives. Iriuchijima and Ogawa reported[18] that the yeast reduction of methyl 4-amino-*N*-benzyloxycarbonyl-2-oxobutanoate having a structure similar to that of our substrate **3** gave (*S*)-α-hydroxyester in 49% ee, which suggests that it is an anti-Prelog product. Hence, it is difficult to predict the stereochemical course in the baker's yeast reduction of an α-oxoester such as **3**. Fortunately, **3** gave the desired (*R*)-hydroxyester (*R*)-**4** as shown in Table I. The run with 1 g of the substrate **3** gave (*R*)-**4** in 34% chemical yield with 54% ee (entry 2).

With baker's yeast reduction of β-oxoesters, the optical purity of the product varies with the size of the ester moiety.[19] To improve the ee in the α-oxoester reduction, we prepared a series of esters, *i.e.* methyl, propyl and butyl ester (**11a**—**c**), and exposed them to baker's yeast

(entries 1, 3 and 4). No improvement of the optical purity was observed. However, in scaling up the run to 40 g of **3**, a slight improvement in optical purity was observed when the substrate was added to the fermenting suspension over a period of 10 h. (*R*)-**4** was obtained in 46% chemical yield with 61% ee (entry 5). This ester was also used in the synthesis of CV-5975 (**1**) as shown in Chart 1.

In summary, we established a procedure using chemoenzymatic processes, lipase hydrolysis and baker's yeast reduction, to prepare the new angiotensin converting enzyme inhibitor CV-5975 (**1**). This new route can be used to prepare **1** in sufficient quantities for biological investigations.

Experimental

All melting points were determined on a Yanagimoto micro melting point apparatus (a hot stage type) and are uncorrected. The infrared (IR) spectra were recorded with a Hitachi 260-10 spectrophotometer. The proton nuclear magnetic resonance (^{1}H-NMR) spectra were recorded on Varian EM-360, EM-390 and XL-100A instruments in the indicated solvents. Chemical shifts are reported as δ-values relative to tetramethylsilane (TMS) as an internal standard. Mass spectra (MS) were obtained on a JEOL JMS-01SC mass spectrometer. The $[\alpha]_D$ values were determined in the indicated solvents on a JASCO DIP 181 4-4822. HPLC analyses were performed on a Shimadzu LC-5A instrument equipped with an SPD-2A detector operating at 254 nm. Peak areas were calculated by using a Shimadzu C-R3A Chromatopac.

Reactions were run at room temperature unless otherwise noted, and followed by thin-layer chromatography (TLC) on Merck Silica gel F_{254} and RP-8F_{254S} plates. Standard work-up procedures were as follows. The reaction mixture was partitioned between the indicated solvent and water. The organic extract was washed in the indicated order with water, $NaHCO_3$ solution (aq. $NaHCO_3$), NaOH solution (aq. NaOH) and hydrochloric acid (aq. HCl), then dried over $MgSO_4$, filtered and evaporated *in vacuo*. Chromatographic separation was done on Merck Silica gel 60 using the indicated eluents.

(*RS*)-6-(1-Benzyloxycarbonyl-4-piperidyl)-2-hydroxyhexanoic Acid ((*RS*)-7)——A solution of NaOH (18 g) in water (200 ml) was added to a solution of (*RS*)-**4** (114 g) in EtOH (100 ml) over a period of 15 min. The mixture was stirred for 20 min, then water (500 ml) was added. The mixture was acidified with conc. HCl and worked up (hexane–AcOEt (1 : 1); water). The oily residue was crystallized from Et_2O to give (*RS*)-**7** (67 g, 64%) as colorless crystals. mp 90—94 °C. IR ν_{max}^{Nujol} cm^{-1}: 3500 (OH), 1730, 1650 (C=O). ^{1}H-NMR ($CDCl_3$) δ: 1.0—1.9 (12H, m), 2.5—3.1 (3H, m), 3.9—4.4 (3H, m), 5.1 (2H, s, CH_2Ph), 7.0 (1H, br, OH), 7.3 (5H, s, phenyl protons). *Anal.* Calcd for $C_{19}H_{27}NO_5$: C, 65.31; H, 7.79; N, 4.01. Found: C, 65.18; H, 7.92; N, 4.04.

Enzymatic Hydrolysis of Ethyl (*RS*)-6-(1-Benzyloxycarbonyl-4-piperidyl)-2-hydroxyhexanoate (Fig. 1)——A mixture of (*RS*)-**4** (5 g), lipase M-AP10 (600 mg) and 0.1 M phosphate buffer (260 ml) was vigorously stirred at 30 °C. After being stirred for 1.5 h, an aliquot (20 ml) of the reaction mixture was taken out, acidified with 1 N aq. HCl and extracted with AcOEt. To determine the conversion (%) of hydrolysis, a small portion of the extract was concentrated *in vacuo* and the residue was analyzed by HPLC under the following conditions: column (Zorbax ODS, 0.46 × 25 cm), mobile phase (water : MeOH = 1 : 5); flow rate (0.3 ml/min) and retention times (20 min for the α-hydroxyester **4** and 14 min for the α-hydroxyacid **7**). The main portion of the extract was washed with aq. $NaHCO_3$ and water, dried over $MgSO_4$ and concentrated *in vacuo* to give the unreacted α-hydroxyester **4**. This alcohol was converted in the usual manner[11] to the corresponding (*S*)-MTPA ester **8**, which was analyzed by HPLC under the following conditions: column (Zorbax ODS, 0.46 × 25 cm); mobile phase (water : MeOH = 1 : 8); flow rate (0.3 ml/min) and retention times (29 min for (*R*),(*S*)-**8** and 32 min for (*S*),(*S*)-**8**). Monitoring of the reaction as described above was repeated after the mixture had been stirred for 3, 4, 5, 6 and 8 h.

Hydrolysis of (*RS*)-**4** (5 g) with lipase PN (25 mg) was carried out similarly to the case of lipase M-AP10, and determination of the relation between conversion (%) and % ee by HPLC was done in the same manner as above.

Stereochemical Assignment of the Remaining Ester in Enzymatic Hydrolysis——The α-hydroxyester **4** (100 mg) with 93% ee, which was obtained by stopping the hydrolysis at point A (lipase PN, 86% conversion) in Fig. 1, was allowed to react with MsCl as described previously[1c] to give the mesylate **5** (115 mg, 95%). A mixture of the mesylate **5** (30 mg) and **2** (51 mg) was heated at 90 °C for 20 h. After cooling, the reaction mixture was worked up (AcOEt; 5% phosphoric acid, water) to give the diester **6** (38 mg, 86%). This diester **6** was analyzed by HPLC under the following conditions: column (Zorbax ODS); mobile phase (water: MeOH = 1 : 8); flow rate (0.3 ml/min); retention times (26 min for (*R*),(*R*)-**6** and 21 min for (*R*),(*S*)-**6**). HPLC analysis showed that the main peak was identical with (*R*),(*S*)-**6** and the ratio of the peak areas of (*R*),(*S*)-**6** and (*R*),(*R*)-**6** was 96.5 : 3.5.

CV-5975 (1)——A mixture of (*RS*)-**4** (40 g), lipase PN (40 mg) and 0.1 M phosphate buffer (1.2 l) was stirred vigorously at 30 °C for 3.5 h. The mixture was acidified with conc. HCl and worked up (AcOEt; water, aq. $NaHCO_3$, water) to give (*R*)-**4** (14.6 g, 37%) in 64% ee. This alcohol (7 g) was converted to the mesylate **5** (8.2 g, 97%) in the

usual manner. $[\alpha]_D^{23.5}$ +15.0° (c=0.52, MeOH). A mixture of this mesylate **5** (8.0 g) and **2** (13.5 g) was heated at 90 °C for 29 h. After cooling, the reaction mixture was worked up (AcOEt; 5% phosphoric acid; water). The oily residue was dissolved in a mixture of oxalic acid (4 g) and Et_2O (50 ml), and the solution was diluted with petroleum ether (200 ml). After cooling, the supernatant layer was removed by decantation, and AcOEt (300 ml) and aq. $NaHCO_3$ (150 ml) were added to the precipitate. The resulting mixture was shaken thoroughly and worked up (AcOEt; water) to give the diester **6** (10 g, 60% de) as an oil. Then 30% HBr–AcOH (30 ml) was added to a solution of this diester **6** in AcOH (15 ml). The mixture was allowed to stand for 2 h and diluted with Et_2O (250 ml). After standing, the supernatant layer was removed by decantation. The residue was dissolved in 1 N aq. NaOH (130 ml). After standing for 1 h, the solution was weakly acidified with AcOH and purified by MCI gel (Mitsubishi Chemical Industries, Ltd., CHP20p, 150—300 μ) chromatography (water : MeOH = 1 : 1). The eluate was concentrated (*ca.* 5 ml) *in vacuo* and allowed to stand overnight. The deposited crystals were collected by filtration to give **1** (2.05 g, 25% based on (*R*)-**4**), which was identical with CV-5975 prepared previously.[1c]

Swern Oxidation of the α-Hydroxyester——The $NaHCO_3$-washing in the enzymatic hydrolysis of (*RS*)-**4** (40 g) with lipase PN was acidified with conc. HCl and worked up (AcOEt; water) to give the (*S*)-α-hydroxyacid (*S*)-**7** (23.5 g, 63%). A mixture of this (*S*)-**7** (22 g), EtOH (50 ml), *p*-toluenesulfonic acid (1.0 g) and toluene (300 ml) was heated at 80—90 °C overnight. After cooling, the mixture was worked up (AcOEt; aq. $NaHCO_3$, water) to give (*S*)-**4** (24 g, 99%) in 32% ee.

A solution of dimethyl sulfoxide (DMSO) (4.45 g) in CH_2Cl_2 (20 ml) was added to a solution of oxalyl chloride (2.3 ml) in CH_2Cl_2 (40 ml) at −65 °C (acetone–dry ice bath) over a period of 10 min, and the mixture was stirred at −60 °C for 10 min. A solution of (*S*)-**4** (5 g, 32% ee) in CH_2Cl_2 (50 ml) was added over a period of 10 min, and the resulting mixture was stirred at −60 °C for 20 min. Diethylisopropylamine (10.3 g) was added over a period of 10 min, and then the acetone–dry ice bath was removed. When the temperature reached −30 °C, 1 N aq. HCl (80 ml) was added to the mixture. After being stirred for 30 min, the mixture was worked up. The residue was purified by silica gel column chromatography (hexane : AcOEt = 4 : 1—2 : 1) to give **3** (2.6 g, 52%)[20] as an oil, which was identical with **3** prepared previously.[1c]

Hydroxy Group Inversion of (*S*)-4 by Kellogg's Method——A mixture of (*S*)-**4** (10 g, 32% ee), lipase PN (20 mg) and 0.1 M phosphate buffer (300 ml) was stirred for 3 h. The mixture was acidified with conc. HCl and extracted with AcOEt (300 ml). The AcOEt layer was washed with aq. $NaHCO_3$ (150 ml). The aqueous layer was acidified with conc. HCl and worked up (AcOEt; water) to give (*S*)-**7** (3.8 g, 41%). This (*S*)-**7** was converted to the ethyl ester in the same manner as described above to give (*S*)-**4** (3.7 g) in 74% ee. This ester (*S*)-**4** (1.0 g) was converted to the mesylate (*S*)-**5** (1.2 g, 99%). $[\alpha]_D^{24.5}$ −16.6° (c=0.5, MeOH).

A solution of propionic acid (0.32 g) in MeOH (4 ml) was added to a solution of Cs_2CO_3 (0.47 g) in MeOH (12 ml). After being stirred for 30 min, the mixture was concentrated *in vacuo*. Toluene (20 ml) was added to the residue, and the mixture was evaporated *in vacuo*. After repeated addition of toluene followed by evaporation (several times), the resulting white powder (CH_3CH_2COOCs) was mixed with (*S*)-**5** (1.2 g) obtained above and *N*,*N*-dimethylformamide (DMF) (30 ml). The mixture was heated at 90 °C for 1 h and worked up (AcOEt; 0.1 N aq. HCl, aq. $NaHCO_3$, water). The oily residue was subjected to silica gel column chromatography (hexane : AcOEt = 3 : 1) to give (*R*)-**9** (1.07 g, 94%) as an oil. IR ν_{max}^{neat} cm^{-1}: 1745, 1700 (C=O). $[\alpha]_D^{24.5}$ +12.2° (c=0.74, MeOH). ^{1}H-NMR ($CDCl_3$) δ: 1.15 (3H, t, J=7 Hz, CH_3), 1.25 (3H, t, J=7 Hz, CH_3), 0.7—2.2 (13H, m), 2.4 (2H, q, J=7 Hz, $COCH_2$), 2.7—3.0 (2H, m, NCH×2), 3.9—4.4 (4H, m, OCH_2, NCH×2), 4.95 (1H, t, J=6 Hz, OCHCOO), 5.15 (2H, s, $C\underline{H}_2Ph$), 7.3 (5H, s, phenyl protons).

Then 1 N aq. NaOH (10 ml) was added to a solution of (*R*)-**9** (1.05 g) in EtOH (10 ml) over a period of 10 min. The resulting mixture was stirred for 30 min, acidified with conc. HCl and worked up (AcOEt; water) to give (*R*)-**7**. This (*R*)-**7** was esterified to (*R*)-**4** (0.82 g, 92%) in a usual manner using EtOH and TsOH. The ee value of this (*R*)-**4** was 70% as determined by HPLC analysis.

Ethyl (*S*)-2-Benzoyloxy-6-(1-benzyloxycarbonyl-4-piperidyl)hexanoate ((*S*)-10)——Benzoyl chloride (0.09 ml) was added to a solution of (*S*)-**4** (0.2 g, 74% ee) in pyridine (2 ml) at 0 °C, and the mixture was stirred at 0 °C for 30 min. After addition of water (1 ml), the mixture was stirred for 1 h and worked up (AcOEt; 1 N aq. HCl, aq. $NaHCO_3$, water) to give (*S*)-**10** (0.25 g, 98%) as an oil. $[\alpha]_D^{24}$ −6.0° (c=0.65, MeOH). IR ν_{max}^{neat} cm^{-1}: 1750, 1720, 1695 (C=O). ^{1}H-NMR ($CDCl_3$) δ: 1.25 (3H, t, J=7 Hz, CH_3), 0.7—2.3 (13H, m), 2.7 (2H, t, J=15 Hz, NCH×2), 3.9—4.4 (4H, m, NCH×2, OCH_2), 5.1 (2H, s, $C\underline{H}_2Ph$), 5.2 (1H, t, J=7 Hz, OCHCOO), 7.2—8.3 (10H, m, phenyl protons).

Mitsunobu Reaction of (*S*)-4——A mixture of (*S*)-**4** (0.7 g, 74% ee), benzoic acid (0.45 g), triphenyl phosphine (0.97 g), diethyl azodicarboxylate (0.65 g) and tetrahydrofuran (THF) (15 ml) was refluxed overnight. The mixture was concentrated *in vacuo* and the residue was subjected to silica gel column chromatography (hexane : AcOEt = 4 : 1—2 : 1) to give (*R*)-**10** (0.38 g, 52%) as an oil. $[\alpha]_D^{25}$ +5.8° (c=0.4, MeOH).

α-Oxoesters (11a—c)——A mixture of (*RS*)-**7** (7 g), MeOH (20 ml), *p*-toluenesulfonic acid (0.3 g) and toluene (100 ml) was heated at 80—90 °C overnight. After cooling, the reaction mixture was worked up (AcOEt; aq. $NaHCO_3$, water) to give methyl 6-(1-benzyloxycarbonyl-4-piperidyl)-2-hydroxyhexanoate (*RS*)-**12a** (6.6 g, 98%) as an oil. IR ν_{max}^{neat} cm^{-1}: 3430 (OH), 1735, 1690 (C=O). ^{1}H-NMR ($CDCl_3$) δ: 0.7—2.0 (13H, m), 2.5—2.95 (2H, m,

NCH × 2), 2.95—3.15 (1H, br d, OH), 3.75 (3H, s, CH_3), 3.9—4.4 (3H, m, NCH × 2, CHCOO), 5.1 (2H, s, $C\underline{H}_2Ph$), 7.3 (5H, s, phenyl protons).

The propyl (**12b**) and butyl (**12c**) esters were prepared similarly using PrOH and BuOH, respectively.

(*RS*)-**12b**: Yield 80%. IR ν_{max}^{neat} cm^{-1}: 3450 (OH), 1730, 1690 (C=O).

(*RS*)-**12c**: Yield 73%. IR ν_{max}^{neat} cm^{-1}: 3450 (OH), 1730, 1695 (C=O).

The α-hydroxyesters (**12a—c**) prepared above were converted to the corresponding α-oxoesters (**11a—c**) by Swern oxidation in a manner similar to that described for the preparation of **3**.

11a: Yield 40%. IR ν_{max}^{neat} cm^{-1}: 1730, 1690 (C=O). MS *m/z*: 361 (M^+). ^{1}H-NMR ($CDCl_3$) δ: 0.7—2.0 (11H, m), 2.5—3.0 (4H, m), 3.85 (3H, s, CH_3), 3.95—4.4 (2H, m, NCH × 2), 5.1 (2H, s, $C\underline{H}_2Ph$), 7.4 (5H, s, phenyl protons).

11b: Yield 45%. IR ν_{max}^{neat} cm^{-1}: 1725, 1695 (C=O). MS *m/z*: 389 (M^+).

11c: Yield 75%. IR ν_{max}^{neat} cm^{-1}: 1725, 1695 (C=O). MS *m/z*: 403 (M^+).

Baker's Yeast Reduction of the α-Oxoesters (Table I)——Entries 1—4: A mixture of baker's yeast (Oriental dry yeast, 10 g), sucrose (20 g) and water (100 ml) was stirred at 30 °C for 10 min. A solution of an α-oxoester (1.0 g) in EtOH (3 ml) was added to the mixture and the resulting mixture was stirred at 30 °C. After 16 h, sucrose (5 g) was added to the mixture and the stirring was continued for a further 30 h. After addition of AcOEt (100 ml), the mixture was filtered through a Celite pad. The AcOEt layer was separated, and the aqueous layer was extracted with AcOEt (100 ml). The extracts were combined and washed with water, dried over $MgSO_4$ and concentrated *in vacuo*. The residue was subjected to silica gel column chromatography (hexane : AcOEt = 2 : 1) to give the corresponding α-hydroxyester as an oil, the ee value of which was determined by the same method as described in the case of lipase hydrolysis.

Entry 5: A mixture of baker's yeast (200 g), sucrose (100 g) and water (2.5 l) was stirred for 10 min. A solution of **3** (40 g) in EtOH–water (3 : 1, 200 ml) and a solution of sucrose (200 g) in water (1 l) were added simultaneously over a period of 10 h. After the addition was complete, the stirring was continued for 38 h, and the mixture was filtered through a Celite pad. The filtrate was extracted with AcOEt–petroleum ether (2 : 1, 750 ml × 2). Insoluble materials were washed with EtOH (1 l × 3), and the EtOH solution was concentrated *in vacuo*. The residue and the extract were combined, and the resulting organic solution was washed with water, 0.1 N aq. NaOH (300 ml) and water, dried over $MgSO_4$ and concentrated *in vacuo*. The residue was purified on silica gel (hexane : AcOEt = 2 : 1) to give (*R*)-**4** (18.4 g, 46%) with 61% ee.

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References and Notes

1) *a*) K. Itoh, M. Kori, Y. Inada, K. Nishikawa, Y. Kawamatsu and H. Sugihara, *Chem. Pharm. Bull.*, **34**, 1128 (1986); *b*) *Idem, ibid.*, **34**, 2078 (1986); *c*) *Idem, ibid.*, **34**, 3747 (1986).
2) The optical purity of **2** was determined by HPLC analysis of the corresponding (*S*)-MTPA amide on Zorbax SIL (hexane : THF : MeOH : H_2O = 40 : 1 : 0.1 : 0.01).
3) Initially we tested the possibility of S_N2-inversion reaction in this system using **2** and ethyl 2-mesyloxylactate. The reaction of (*S*)-mesyloxylactate with **2** (100—110 °C, 3 h) gave a single product which was identical with the (*R*),(*R*)-diester[1a] on TLC (hexane : AcOEt = 2 : 1), while the reaction of the racemic mesyloxylactate provided a diastereomeric mixture (*ca.* 1 : 1).
4) Successful S_N2-inversion reactions of chiral α-sulfonates have been reported: *a*) V. F. Effenberger, U. Burkard and J. Willfahrt, *Angew. Chem.*, **95**, 50 (1983); *b*) H. Urbach and R. Henning, *Tetrahedron Lett.*, **25**, 1143 (1984); *c*) R. M. Scott, Japan. Patent 59-59120 (1977) [*Chem. Abstr.*, **87**, 67990*n* (1977)].
5) Salts with quinidine, cinchonine, (*R*)-(+)-α-methylbenzylamine and dehydroabiethylamine failed to crystallize.
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7) Lipase A and pancreatin are inactive in this hydrolysis.
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10) In the absence of enzymes, no hydrolysis was observed.
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20) The chemical yield of the product **3** was slightly reduced by dimerization during the purification by silica gel chromatography. Afterwards, it was found that when silica gel was treated with AcOH before use (a mixture of silica gel (700 g), AcOH (20 ml) and hexane–CH_2Cl_2 (1 : 1, 500 ml) was evaporated thoroughly *in vacuo*), dimerization of **3** was suppressed.