Tetrahedron Vol. 49, No. 26, pp. 5805-5816, 1993
Printed in Great Britain

0040-4020/93 $6.00+.00

Reductive Biotransformation of Carbonyl Compounds--- Application Of Fungus, *Geotrichum sp.*G38 in Organic Synthesis

Gu Jian-Xin, Li Zu-Yi, Lin Guo-Qiang*
Shanghai Institute of Organic Chemistry, Chinese Academy of Sciences, 345 Ling Ling Lu, Shanghai 200032, China

(*Received in Japan* 8 *March* 1993)

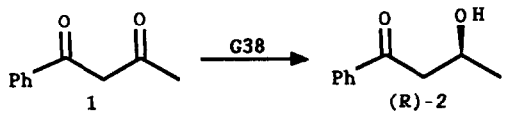
ABSTRACT: The microbial transformation of 2- and 3-oxo esters and diketones with *Geotrichum sp.***G38** and its application to the syntheses of the key intermediates of several bioactive compounds such as (R)-denopamine **8**, (R)-fluoxetine **11** and (2S,3R)-sitophilate **14** were described.

There is a continuing need for the new direct methods in the synthesis of optically active compounds, and the great advances have been made in this area.[1] Among many of asymmetric synthesis, transformation catalyzed by enzymes and microorganisms(biotransformation) is now well-recognized as valuable tool in preparation of chiral intermediates. Reduction of carbonyl groups is probably the most thoroughly studied and exploited because of its utilities.[2]

Baker's yeast, *Saccharomyces cerevisiae*, has attracted more interest and plays an important role because it is inexpensive, versatile, easily available and it can be grown without the assistance of a microbiologist. It has been known that baker's yeast can convert various classes of compounds, especially carbonyl-containing substrates into the corresponding optically active alcohols,[3] usually with S configuration. For the synthesis of various kinds of natural products and physiologically active compounds, it is desirable to find a microorganism which has the ability to perform reduction with opposite enantioselectivity compared with baker's yeast.

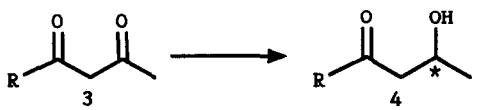
1. Screening and reduction of 1,3-diketones

Screening of a microorganism was carried out using 1-phenyl-1,3-butanedione **1** as a substrate and we found that *Geotrichum sp.***G38**, a fungus isolated from a soil sample, could give the product **2** in which only one carbonyl group was regioselectively reduced with R configuration in 95% optical yield. Encouraged by this result, reaction of other compounds



with this strain was examined. In all cases (R)-alcohols were obtained. Table 1 shows that the reduction of β-diketones **3** with **G38** yielded (R)-**4**, whereas baker's yeast produced (S)-**4**.[4] These results are very useful because **4** can be converted into syn or anti-1,3-diols, the subunits of many natural products, by the conventional methods.[5]

Table 1. Bioreduction of β-diketones **3** with baker's yeast and **G38**



Entry	R	with baker's yeast				with G38			
		time(h)	yield(%)	ee[a]	conf.[b]	time(h)	yield(%)	ee[a]	conf.[b]
a	Me	36	16	80	S	12	21	92	R
b	Et	48	23	88	S	15	38	95	R
c	Pr	48	32	75	S	18	53	99	R
d	Bu	48	42	91	S	20	77	99	R
e	Pen	60	46	86	S	24	75	47	R

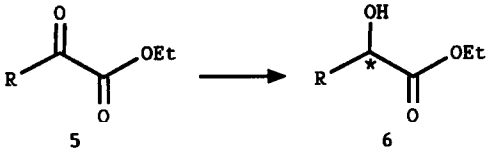
[a]The optical purities were determined by HPLC(CLC-Sil,n-hexane:ethyl acetate=9:1) analysis of their corresponding MTPA-esters. [b]The absolute configurations were assigned by comparison with literature data.

2. Reduction of α-oxo esters with G38

Optically active α-hydroxy esters are important intermediates for chiral syntheses of bioactive substances such as pheromones[6] and antibiotics.[7] Using **G38** at room temperature, we discovered that the α-oxo esters **5** can be reduced effectively to the corresponding α-hydroxyesters **6** in excellent optical yield. The configuration of the products was S when R=alkyl whereas it was R when R=aryl. The results were summarized in Table 2.

In the reduction of α-oxo esters with this fungus, it seems that the outcome of induced stereoselectivity was independent on the bulkiness of R. In the case of **5j**, it might be due to the existence of phenol group which is toxic to the microorganism, the rate of conversion was slow, and only 10% substrate was transformed into the corresponding hydroxy product **6j** over 60 h incubation in 39% optical yield. The satisfactory result

Table 2. Bioreduction of 5 with G38

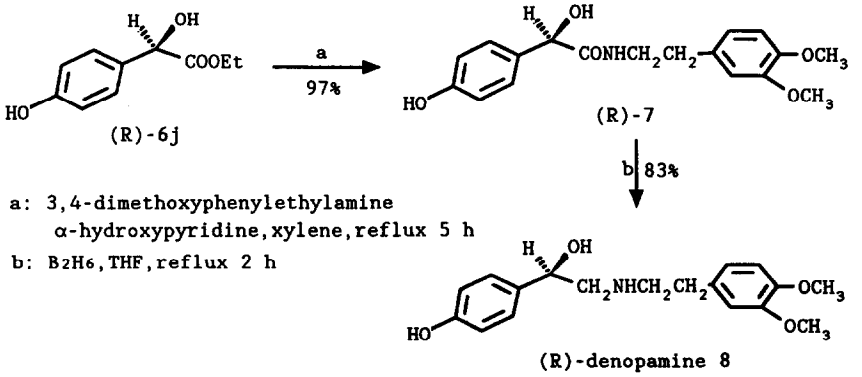


5 6

	R	time(h)	yield(%)	$[\alpha]_D$	ee(%)	conf.
a	CH_3	4	47	-8.43^0(c 1.2,EtOH)	90[a]	S
b	C_2H_5	5	47	-6.55^0(c 1.1,EtOH)	83[b]	S
c	i-C_3H_7	4	42	-6.4^0(c 1.25,CCl_4)	92[c]	S
d	n-C_4H_9	6	40	-5.5^0(c 1.2,EtOH)	95[c]	S
e	i-C_4H_9	4	36	-5.84^0(c 1.0,EtOH)	92[c]	S
f	n-C_5H_{11}	4	35	-5.8^0(c 1.3,EtOH)	88[d]	S
g	$PhCH_2CH_2$	24	48	21.7^0(c 0.9,$CHCl_3$)	98.5[e]	S
h	Ph	16	76	-105.8^0(c 2.5,EtOH)	100[f]	R
i	1-naphthyl	8	95	-146.7^0(c 2.4,$CHCl_3$)	96[g]	R
j	p-HOC_6H_4	60	10	-31.6^0(c 1.2,CH_3COCH_3)	39[h]	R
k	p-$AcOC_6H_4$	10	93	-81.8^0(c 1.1,CH_3COCH_3)	99.5[h]	R

[a] Based on $[\alpha]_D=-9.36^0$(EtOH).[9] [b] Based on $[\alpha]_D=-7.88^0$(c 1.46,EtOH).[10] [c] Determined by HPLC analysis of MTPA esters. [d] Based on $[\alpha]_D=-6.6^0$(c 7.1, MeOH).[11] [e] Based on $[\alpha]_D=-22.1^0$(c 1.0,$CHCl_3$).[12] [f] Based on $[\alpha]_D=-104^0$(c 1.75, EtOH).[13] [g] Based on $[\alpha]_D=153^0$(c 2.68,$CHCl_3$).[8] [h] Optical purity was determined after convertion of the ester into the corresponding acid with KOH-EtOH at 0^0C, the acid $[\alpha]_D$(max.)$=144^0$(c 0.6,H_2O).[14]

could be obtained from the corresponding acetate **5k**. Under the same condition, the conversion was completed in 10 h, giving **6j** with 99.5%ee in 93% yield.

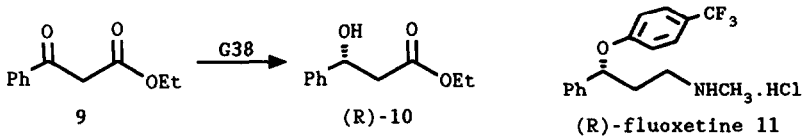


Amidation of (R)-**6j** using α-hydroxypyridine as catalyst followed by reduction with borane, (R)-denopamine **8**, a new selective β_2-agonist was obtained in a total yield of 74.9%. This amine is important for the treatment of congestive heart failure without promotion of increased myocardial oxygen consumption or heart rate.[15] To our knowledge, this chemical-enzymatic method is the second stereospecific route to such drug. The first asymmetric synthesis of **8** were developed by Corey,[16] in which (R)-denopamine **8** and its enantiomer was synthesized by using the CBS[17] enantioselective catalytic reduction process in 60% total yield over six steps.

3. Reduction of β-oxo esters with G38

Reduction of β-oxo esters by using baker's yeast is probably one of the most extensively studied topics.[3] They can be also converted by *Geotrichum sp.***G38** resulting in the alcohols of opposite stereochemistry. For example, reduction of **9** with **G38** yielded (R)-**10**, whereas baker's yeast produced the antipode (S)-form.

Based on above two biotransformations by **G38** and baker's yeast, both (R) and (S)-fluoxetine **11** were produced in five steps in overall yields of about 60%.[18] (R)-fluoxetine, the serotonin-uptake inhibitor, is one of the most exciting new therapeutic agents.[19]

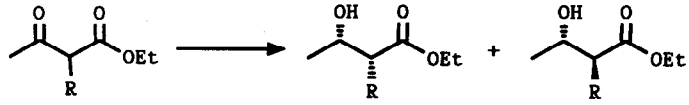


9 (R)-10 (R)-fluoxetine 11

Different from using pure enzyme, bioreduction of racemic α-substituted β-keto esters with baker's yeast furnished a mixture of syn and anti aldol products. **G38** also showed simillar enantio- and diastereoselectivity. For example, reduction of ethyl α-substituted acetoacetate **12** with resting cells of **G38** gave a mixture of syn and anti **13**. In most cases the ratios of anti/syn are about 3:1. If the incubation was performed under anaerobic condition(N_2),[20] the syn isomers became the major. The results are illustrated in Table 3.

We found that the rates of conversion were slower and the stereoselectivities were higher under anaerobic condition than under aerobic condition.

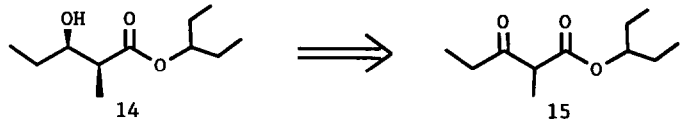
Table 3. Reduction of **12** with **G38** under aerobic and anaerobic conditions



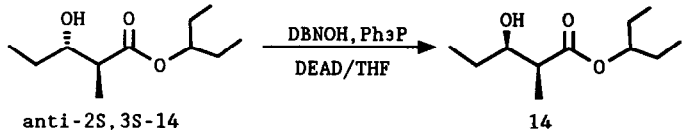
entry	R	aerobic condition time(h)	yield(%)	anti:syn[a]	anaerobic condition time(h)	yield(%)	anti:syn[a]
a	Me	48	78	75 : 25	72	80	40 : 60
b	Et	72	88	68 : 32	72	89	32 : 68
c	Bu	48	84	61 : 39	60	87	37 : 63
d	Allyl	72	91	80 : 20	84	83	25 : 75
e	Bn	60	98	98 : 2	72	99	20 : 80

[a]The ratio was determined by capillary GLC(FFAP, 50m, 150°C+5°C/min)

Sitophilate **14**, the aggregation pheromone of a grainary weevil *sitophilus granarius*,[21] is a syn α-substituted β-hydroxy ester. It could be prepared by microorganism-mediated reduction of the corresponding oxo ester **15** as follows.



At first, **15** was subjected to the microbial reduction with **G38** under anaerobic conditions. To our disappointment, almost equal amount of syn and anti **14** was obtained. We therefore treated **15** with resting cells of **G38** under aerobic condition, to obtain 56% yield of anti-(2S,3S)-**14**(anti: syn=97.4:2.6), which then could be easily transformed to (2S,3R)-**14** by Mitsunobu inversion.[22] If **G38** was immobilized on silicone polymer prepared according to a modified Oriel's method,[23] the anti-(2S,3S)-**14**



became nearly single product(anti: syn=99.2:0.8). These results are better than those obtained by *Pichia farinosa* IAM 4682, a yeast which has an unusual aptitude, so called anti-prelog's rule selectivity,[24] discovered by Ohta et al.[25] They reported that (2S,3S)-**14** was obtained as the major product (63% yield, 43% de.) *via* the reaction of enol ester of

15 with growing cells of this yeast.

4. Reduction of furfuryl carbonyl compounds with G38

The possibility of transformation of other functional compounds was examined. Fermentation of α-acylfurans **16** with **G38** furnished the corresponding chiral alcohols, furfuryl carbinols **17**, which can be conveniently transformed into the hydropyranones by the established oxidative techniques,[26] and this pyranones would be well suited as starting materials for the preparation of a variety of biologically important molecules.[27] Table 4 summarized the results of the conversion of **16** into **17**. It was revealed that **17c**(R=Pr) was a good substrate.

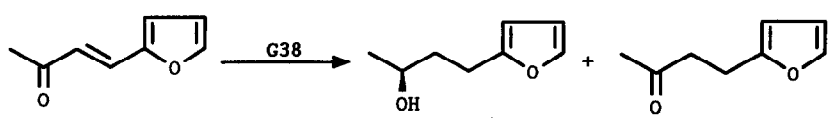
Table 4. Bioreduction of α-acylfurans **16** with **G38**

16 17

	R	time(day)	yield(%)	$[\alpha]_D$(c,ethanol)	ee(%)
a	Me	2	60	-15.4°(1.2)[a]	74[b]
b	Et	2	53	9.2°(1.1)[a]	46[c]
c	Pr	2	44	24.7°(1.4)	100[d]
d	Bu	3	34	13.4°(0.9)	64[d]
e	Pentyl	4	22	-15.4°(1.1)	66[d]
f	Ph	4	26	23.7°(1.0)[a]	100[d]

[a] Measured in $CHCl_3$. [b] Based on (R)-form, $[\alpha]_D$=20.8°(c 1.27,$CHCl_3$).[28] [c] Based on calculated $[\alpha]_D$(max.)=19.3°(c 1.0,$CHCl_3$).[29] [d] Determined by HPLC (CLC-Sil, hexane:ethyl acetate=10:1) analysis of their corresponding MTPA-ester.

Another good example is furfuralacetone **18**. The microbial reduction of α,β-unsaturated ketone **18** with resting cells of **G38** gave a mixture of saturated alcohol (S)-**19** and saturated ketone **20** in 86% and 10% yield, respectively.

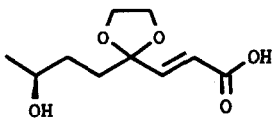


18 (S)-19 20

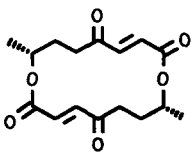
The optical purity of the major product (S)-**19** was over 98%, determined by ^1H NMR analysis of the corresponding acetate in the presence of $Eu(hfc)_3$.

7-(S)-hydroxy-4,4-ethylenedioxy-2-octenoic acid **21** was synthesized from

(S)-**19** via Petrint's procedure.[30] Hydroxy acid **21** is a protected building block of the C-8 subunit of (-)-pyrenophorin **22**,[31] which is a naturally occurring antifungal macrodiolide produced by the plant pathogenic fungus *pyrenophora avenae*. By using the Mitsunobu reaction,[32] the chiral intermediate (S)-**21** could be easily converted into this macrodiolide by the known dimerizing cyclization procedure.[33] In comparison with the previous approaches,[34] this microbial-chemical method is more practical



21



22

and effective.

Experimental Section

General Methods

Melting points were uncorrected. IR spectra were determined on an IR-440 spectrometer. The ^1H NMR spectra were recorded on an EM-360 and an XL-200 spectrometers. Mass spectra(MS) were recorded on a Finnigan 4021 instrument. Optical rotations were measured with a Perkin-Elmer 241 polarimeter. HPLC was analyzed with a Shimadzu LC-6M. GC was analyzed with a HP 5880A instrument.

Cultivation of *Geotrichum sp.*G38

The following culture medium was used: glucose(10%), yeast extract(1%), urea(0.1%). *Geotrichum sp.*G38 was inoculated to the sterilized medium(100 ml) in 500 ml-shaking flask and the flask was shaken(180 rpm) for 2 days at 28°C. This seed culture was added to the medium(1000 ml), which was shaken at 28°C, after 24h growth, the culture was filtered and the mycelium (80g) was washed several times with 0.8% NaCl solution.

General Procedure for Microbial Reduction of Diketones 3 with G38

Wet mycelium(8g) was suspended in 250 ml-flasks containing glucose solution(5%,50ml) and diketones 3a-e(1 mmol), the flask was shaken at 180 rpm at 28°C. After reaction, the mixture was filtered and the filtrate was extracted with ether. The organic layer was dried($MgSO_4$) and evaporated under reduced pressure. Purification of the crude products by chromatography on silica gel eluting with petroleum ether:ethyl acetate (10:3) afforded (R)-2-hydroxy-4-alkanones 4a-e as colorless oil. The

yields were cited in table 1. As a representative example, **4c**: $[\alpha]_D$= -67.2°(c1.5, $CHCl_3$); ^1H NMR($CDCl_3$),δ:1.05-1.33(m,8H),2.25(t,J=6Hz,2H), 2.40(d,J=6Hz,2H),2.58(s,1H),4.10(m,1H)ppm.

Reduction of 5a-f with G38

The bioconversion conditions were identical with those used for reduction of diketones **3**. For a representative example, **6b**: $[\alpha]_D$=-6.55°(c 1.1, EtOH),IR(film):3300,2975,1735cm^{-1}. ^1H NMR($CDCl_3$),δ:0.95(t,J=7Hz,3H), 1.3(t,J=7Hz,3H),1.4-2.0(m,2H),2.8(s,1H),3.98-4.4(m,1H),4.25(q,J=7Hz,2H) ppm. m/z:132,115,59.

Reduction of 5g-k with G38

Wet mycelium **G38**(10g) was suspended in tap water(50ml), **5g-k**(150mg) were placed and then shaked at 28°C for time cited in table 2. After usual work-up, the optical active **6g-j** were obtained. For a representive example, **6j**: a white solid. mp:128-129°C, IR(KCl):3410,1735,1610,1595, 1510cm^{-1}. ^1H NMR(CD_3COCD_3),δ:1.3(t,J=7Hz,3H),3.4(ws,2H),4.3(q,J=7Hz,2H), 5.25(s,1H),7.3(AB,4H). m/z:196,123,95,77,43.

Synthesis of (R)-7

A solution of **6j**(196mg,1mmol,$[\alpha]_D$=-81.8°(acetone)), α-hydroxypyridine (70mg,0.75mmol) and 2-(3,4-dimethoxyphenyl)ethylamine was refluxed in xylene(3ml) under N_2 for 5h. After cooling, the solution was diluted with ethyl acetate, the organic layer was washed subsequently with dilute hydrochloric acid, 10% $NaHCO_3$ and saturated NaCl. The organic layer was dried over sodium sulfate, filtered and concentrated by rotary evaporation. The crude product was chromatographed on silica gel eluting with petroleum ether: ethyl acetate(1:1) to yield (R)-**7**(321mg,97%). mp:146-147°C,$[\alpha]_D$=-58.9°(c 1.0,acetone),IR(KCl):3340,1640,1615,1595,1520, 1245,1020cm^{-1}, ^1H NMR(CD_3COCD_3),δ:2.75(t,J=7Hz,2H),3.4(m,2H),3.82(s,6H), 4.8(s,2H),4.92(s,1H),6.7-6.9(m,5H),7.27(AB,4H),7.93(m,1H)ppm, HRMS: $C_{18}H_{21}NO_5$, calcd. 331.1414, found 331.1388.

Synthesis of (R)-Denopamine 8

To 1M diborane(in THF)(2ml,2mmol) cooled to 0°C was added dropwise (R)-**7**(160mg,0.5mmol) in dried THF(1ml) under N_2. After addition, the mixture was refluxed for 2h. Then the reaction mixture was cooled in an ice bath and hydrogen chloride gas was introduced. After stirring for another 1.5 h, the reaction mixture was concentrated in reduced pressure and diluted with ethyl acetate. A solid was obtained by filtration, which was dissolved in 5% $NaHCO_3$ and then saturated with brine, the solution was extracted with ethyl acetate. This organic layer was dried with anhydrous potassium carbonate and concentrated. The white solid was then obtained and recrystallized from ethyl acetate-hexane to provide (R)-**8** as a white solid(131mg,83%). mp:164-165°C, $[\alpha]_D$=-28.3°(c 1.1,MeOH)[lit.[35]

mp:167°C dec., $[\alpha]_D$=-27.7°(c 1.0,MeOH)].IR(KCl):2970,1615,1590,1510cm^{-1}, ^{1}H NMR(CD_3COCD_3),δ:2.2(s,1H),2.7-3.0(m,6H),3.4(ws,2H),3.8(s,6H),4.6(m,1H),6.7-6.9(m,5H),7.22(d,J=8Hz,2H)ppm, HRMS:$C_{18}H_{23}NO_4$,calcd.318.1704, found 318.1663.

Bioconversion of 12a-e with G38 under Aerobic Condition

The bioconversion conditions were identical with bioreduction of **3**.

Bioconversion Of 12a-e with G38 under Anaerobic Condition

The bioconversion conditions were identical with above except that nitrogen was bubbled through the suspension of cells before addition of substrates and the flasks were sealed with septum caps.

Preparation of anti-(2S,3S)-14 by Bioconversion of 15 with G38

The washed wet cells(5g) was suspended in tap water(50ml) and oxo ester **15**(150mg) was added. The mixture was shaken at 28°C for 60 h, the reaction was stopped by filtration, followed by ether extraction of the solution. The organic layer was dried and concentrated, the residue was chromatographed on silica gel with petroleum ether:acetone(5:1) to yield anti-2S,3S-**14**(84mg,56%),^{1}H NMR($CDCl_3$),δ:0.9-1.67(m,18H),2.42(m,1H),2.92(s,1H),3.76(m,1H),4.16(m,1H)ppm. The diastereomeric ratio analyzed by GC (FFAP,50× 0.2mm,220°C)was 97.2:2.8.

General procedure for bioreduction of 16 with G38

The procedure is in the same manner as described in reduction of **3**. As a representative example, the data for **17c** is shown below. $[\alpha]_D$=24.7°(c 1.4,EtOH), IR(neat):3350,2960,1470,1050,720cm^{-1}, ^{1}H NMR($CDCl_3$),δ:0.9(t,J=7Hz,3H),1.0-2.1(m,4H),3.1(s,1H),4.5(t,J=7Hz,1H),6.05(s,1H),6.12(s,1H),7.2(s,1H). m/z:140,123,98.

Bioconversion of 18 with G38

Freshed wet cells(30g) were suspended in flask containing tap water (150ml) and substrate **18**(300mg). This flask was shaken for 24 h at 28°C. After usual work-up, the crude product was purified by chromatography on silica gel with petroleum ether:ethyl acetate(10:3) to yield **19**(269mg, 86%); $[\alpha]_D$=-22.6°(c 1.5,$CHCl_3$). IR(neat):3360,2970,1050,720cm^{-1}.^{1}H NMR ($CDCl_3$),δ:1.22(d,J=6.5Hz,3H),1.62-1.93(m,2H),2.43(s,1H),2.75(t,J=7Hz,2H),3.78(m,1H),5.96(s,1H),6.24(s,1H),7.28(s,1H); and **20**(30mg,10%);^{1}H NMR (CCl_4),δ:2.04(s,3H),2.11(t,J=7Hz,2H),2.78(t,J=7Hz,2H),6.02(s,1H),6.28(s,1H),7.30(s,1H)ppm.

Synthesis of 21 from 19

A solution of **19**(210mg,1.5mmol), acetic anhydride(204mg,2mmol) and anhydrous pyridine(0.5ml) was stirred at room temperature over night. The reaction was queched by dilution with water and extracted with ether. The organic layer was washed with dilute hydrochloric acid, cupric sulphate, water, 5% $NaHCO_3$ and brine, respectively, then dried with anhydrous

sodium sulfate. Concentration to dryness provided the corresponding acetate compound(271mg,99%). $[\alpha]_D$=-3.5°(c 1.0,$CHCl_3$).IR(neat):3020,2980, 1730,1370,1040,890cm^{-1}.^1H NMR($CDCl_3$),δ:1.22(d,J=6.5Hz,3H),1.90(m,2H),1.98 (s,3H),2.65(t,J=6.5Hz,2H),4.96(m,1H),6.01(s,1H),6.30(s,1H),7.32(s,1H)ppm. m/z:182,165,161,149,139,123,108,81. The optical purity judged by 200MHz ^1H NMR in the presence of Eu(hfc)$_3$ was over 98%.

The above acetate compound(183mg,1mmol) was dissolved in dichloromethane (20ml). The solution was stirred and PCC(1.08g,5mmol) was added. The stirring was continued at room temperature over night and then refluxed for 12h. After cooling, the mixture was diluted with ether(20ml) and filtered. The filtrate was washed several times with ether. Removal of the combined solvent under reduced pressure afforded an oil(140mg).

This oil was placed in acetone(5ml) and cooled to 0°C. Jones reagent (0.25ml of 8M,2mmol) was added dropwise with vigorous stirring. After 2 h, 2-propanol was added to destroy the excess oxidant. The resulting greenish solution was diluted with ethyl acetate and decanted. The combined organic solution was then successively washed with aqueous 10% hydrochloric acid and water until the organic layer was slight yellow. Drying and concentration in vacuo gave an oil(145mg). The crude oil, which was a mixture of cis /trans isomers, was placed in benzene(1.5ml) and 2-mercaptobenziimidazole (2mg) was added. The resulting solution was stirred at room temperature for 6 h. Ethylene glycol(50mg,0.8mmol) and p-toluenesulfonic acid(2mg) was added. The solution was refluxed for 24 h. After cooling, water was added and the resulting mixture was extracted with ethyl acetate.Drying and concentration provided a yellow oil(136mg).

The above oil was placed in 50% aqueous tetrahydrofuran(1.5ml), lithium hydroxide(52mg,2mmol) was added. After stirring for 20 h, reaction mixture was acidified and extracted with ethyl acetate. Drying and concentration gave a yellow oil, which was purified by flash chromatography(petroleum ether:ethyl acetate:acetic acid=8:2:0.1) to give (S)-**21**(65mg);$[\alpha]_D$=9.6°(c 1.3,$CHCl_3$) [lit.[36] $[\alpha]_D$=9.1°(c 1.3,$CHCl_3$),ee> 99%]. IR(neat):3250,3000,2960,1690,1650,1410,1260,1050,940cm^{-1}.^1H NMR ($CDCl_3$),δ:1.2(d,J=6.5Hz,3H),1.52-2.0(m,4H),3.90(m,4H),4.17(m,1H),6.05(d, J=16Hz,1H),6.9(d,J=16Hz,1H),7.8(brs,2H)ppm. m/z:217(M+1),199,171,143,99, 83,45.

Reference

1. Wong, C.-H., Chemtracts--Organic Chemistry,**1990**, 3, 91.
2. Treilhou, M.; Fauve, A.; Pougny, J.R.; Prome, J.C.; Veschambre, H.,

J. Org. Chem.,**1992**, 57, 3203; Keinan, E.; Sinha, S.C.; Singh, S.P., Tetrahedron, **1991**, 47, 4631; Hsu, C.T.; Wang, N.Y.; Latimer, L.H.; Sih, C.J., J. Am. Chem. Soc., **1982**, 104, 4251.

3. Servi, S., Synthesis, **1990**, 1.
4. Ohta, H.; Ozaki, K.; Tsuchihashi, G., Agric. Biol. Chem., **1986**, 50, 2499; Fauve, A.; Veschambre, H., J. Org. Chem., **1988**, 53, 5215.
5. Evans, D.A.; Chapman, K.T., Tetrahedron Lett., **1986**, 27, 5939.
6. Mori, K.; Sasaki, M.;Tamada, S.; Suguro, T.; Masuda, S., Tetrahedron, **1979**, 35, 1601.
7. Caglioti, L.; Mitisi, D.; Mondelli, R.; Selva, A.; Arcamone, F., Cassinelli, G, Tetrahedron, **1969**, 25, 2193.
8. Brown, H.C.; Chandrasekharan, J.; Ramachandran, P.V., J. Org. Chem., **1986**, 51, 3396.
9. Beckett, A.H.; Hepper, N.J.; Clitherrow, J.W., J. Pharm. Pharmacol., **1963**, 15, 349.
10. Nakamura, K.; Inoue, K.; Ushio, K.; Oka, S.; Ohno, A.,J. Org. Chem., **1988**, 53, 2589.
11. Larchereque, M.; Pitet, Y., Bull. Soc. Chem. Fran., **1989**, 130.
12. Urbach, H.; Henning, R., Tetrahedron Lett., **1984**, 25, 1143.
13. Yamada, T.; Narasaka, K., Chem. Lett., **1986**, 133.
14. Pratesi, P.; Manna, A.L.; Campiglio, A.; Ghislandi, V., J. Chem. Soc., **1958**, 2069.
15. Bristow, M.R.; Hershberger, R.E.; Port, J.D.; Minobe, W.; Rasmussen, R., Mol. Pharmacol., **1989**, 35, 295.
16. Corey, E.J.; Link, J.O.; J. Org. Chem., **1991**, 56, 442.
17. Corey, E.J.; Bakshi, R.K.; Shibata, S.; Chen, C.-P.; Singh, V.K., J. Am. Chem. Soc., **1987**, 109, 7925.
18. Gu, J.X.; Li, Z.Y.; Lin, G.Q., Chin. J. Chem., **1992**, 10, 355.
19. Ankier, S.I.; Prog. Med. Chem., **1986**, 23, 121.
20. Fauve, A.; Veschambre, H., Tetrahedron Lett., **1987**, 28, 5037
21. Phillips, J.K.; Miller, S.P.F.; Andersen, J.F.; Fales, H.M.; Burkholder, W.E., Tetrahedron Lett., **1989**, 28, 6145.
22. Mori, K.; Ishikura, M., Liebigs Ann. Chem., **1989**, 1263.
23. Oriel, P., Enzyme Microb. Technol., **1988**, 10, 518.
24. Shieh, W.R.; Gopalan, A.S.; Sih, C.J., J. Am. Chem. Soc., **1985**, 107, 2993.
25. Sugai, T.; Sakuma, D.; Kobayashi, N.; Ohta, H., Tetrahedron, **1991**, 47, 7237.
26. Ho, T.-L.; Sapp, S.G., Synth. Commun., **1983**, 13, 207.
27. Martin, S.F.; Guino, D.E., J. Org. Chem., **1987**, 52, 5588.
28. Kusakabe, M.; Kitano, Y.; Kobayashi, Y.; Sato, F., J. Org. Chem.,

1989, 54, 2085.

29. Devem, A.; Menge, W.; Ferirga, L., Tetrahedron Lett., **1989**, 30,6427.
30. Petrint, M.; Ballint, R.; Rosini, G.; Marotta, E., Tetrahedron, **1986**, 42, 151.
31. Nozoe, S.; Hirai, K.; Tsuda, L.; Ishibashi, K.; Shirasaka, M.;Grove, J.F., Tetrahedron Lett., **1965**, 4675.
32. Mitsunobu, O., Synthesis, **1981**, 1.
33. Bakuzis, P.; Bakuzis, M.L.F.; Weingartner, T.F., Tetrahedron Lett., **1978**, 2371.
34. Seuing, B.; Seebach, D.; Liebigs Ann. Chem., **1978**, 2044; Mall, R.S.; Pohmakotr, M.; Weidmann, B.; Seebach, D., *ibid*, **1981**, 2272.
35. Kezaki, I.;Umino, N.; Gaino, M.; Aoe, K.; Iwakuma, T.; Ohishi, T., Yakugaku Zasshi, **1986**, 106, 80.
36. Ngooi, T.K.; Scilimati, A.; Guo, Z.W.; Sih, C.J., J. Org. Chem., **1989**, 54, 911.