

Lipase catalysed resolution and microbial reduction for obtaining enantiopure 1-(2-thienyl)alkanols

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Dedicated to Professor Kjell Undheim on the Occasion of his 70th birthday
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Abstract

Efficient methods have been developed for resolving 1-(2-thienyl)alkanols with lipase B from *Candida antarctica* as catalyst. Fermenting cells and cell-free reduction systems of *Geotrichum candidum* (IFO 4597) have also been tested in asymmetric reductions of 1-(2-thienyl)alkan-1-ones.

Keywords : Lipase, resolution, 1-(2-thienyl)alkanols, microbial reduction

Introduction

Kinetic resolution is an efficient method for providing both enantiomers of chiral compounds. The success of a process depends profoundly on the ability of the enzyme to select between the two enantiomers as expressed by the enantiomeric ratio *E*, which is the relative rate of reaction with the two enantiomers.¹ Compared to asymmetric synthesis, kinetic resolution has the benefit that a high enantiomeric excess can be obtained even with a moderate *E*-value. One of the shortcomings of lipase catalysed resolutions is that the maximum yield is limited to 50 %. This can be overcome for instance by performing a dynamic kinetic resolution in which the unreacted enantiomer is continuously racemised.

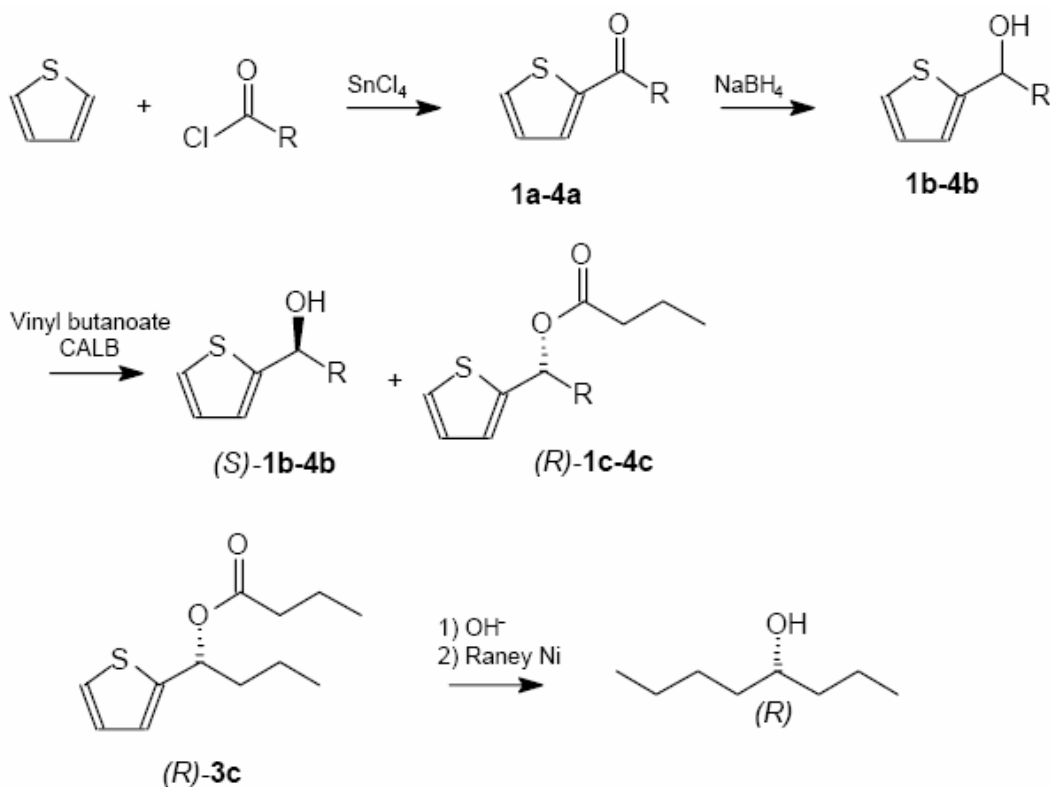
Various micro-organisms have been used for oxidative kinetic resolution of 1-(2-thienyl)ethanol.²⁻⁴ 1-(2-Thienyl)propanol has previously been resolved with *Pseudomonas cepacia* as catalyst with moderate *E*-values (15-30) depending on temperature.⁵ (*R*)-1-(2-Thienyl)ethanol and (*R*)-1-(2-thienyl)propanol have been prepared with ee of 91 % and 88 % respectively using an oxazaborolidine catalyst.⁶ (*R*)-1-(2-Thienyl)ethanol has also been reported

via a Rhodium-catalysed asymmetric hydrosilylation in good yields and 76 % ee.⁷

Results and Discussion

Synthesis of starting compounds

The ketones **2a-4a** were synthesised by a modified Friedel-Crafts acylation of thiophene⁸, using SnCl₄ as the Lewis acid as shown in Scheme 1 (**1a** was purchased). Thiophene was reacted with the appropriate acid chloride and the yield was 80-90 %. The corresponding ketones were reduced with sodium borohydride to give the racemic alcohols in good yields.



Scheme 1. Synthesis of 2-acyl thiophenes, reduction to 1-(2-thienyl)-1-alkanols and lipase catalysed resolutions, R = Me (**1**), Et (**2**), Pr (**3**), Bu (**4**). In order to prove the absolute configuration of **3c**, it was converted to (*R*)-4-octanol.

Lipase catalysis

The four secondary alcohols were resolved in a series of solvents with lipase B from *Candida antarctica* (CALB) as catalyst. Vinyl butanoate was used as acyl donor.

CALB displayed high E-values for all secondary alcohols. We have previously explored the substrate requirements of this enzyme and found that in organic solvents an ethyl group is the largest of the “small” substituent (in the present substrates the “large” substituent is the

thiophene moiety) of a secondary alcohol/ester in order to obtain a high E-value⁹. However, for the present thiophenes, also R = propyl or butyl also gave very high E-values (Table 1).

Table 1. E-values of CALB catalysed kinetic resolution of 1-(2-thienyl)alkanols

	R	Hexane	Benzene	Et ₂ O	Dioxane	Toluene	t.-BuOMe	CCl ₄
1b	Me	204	211	221	167	236	217	146
2b	Et	>500	>500	>500	382	>500	>500	>500
3b	Pr	148	>500	216	176	218	369	349
4b	Bu	196	150	152	73	237	90	66

Stereo preference of resolutions

For both 1-(2-thienyl)ethanol (**1b**) and 1-(2-thienyl)propanol (**2b**), the (*R*)-enantiomer was the faster reacting in the transesterifications, shown by comparing the optical rotation of the remaining alcohol with previously reported values.²⁻⁵ Since no optical rotation has been reported for enantiopure 1-(2-thienyl)butanol (**3b**) or the corresponding butanoate, the ester was hydrolysed and treated with Raney-Ni to give (*R*)-4-octanol¹⁰ with $[\alpha]_D^{20} = +0.25^\circ$. This shows that the (*R*)-butanoate also was formed in this case. This way of proving the configuration is not applicable for 1-(2-thienyl)pentanol (**4b**) since the formed secondary alcohol is achiral. However, based on knowledge of the stereo preference of CALB and the elution order of enantiomeric alcohols and esters (Table 2), it is reasonable to assume that the (*R*)-ester and the (*S*)-alcohol are the products also in the resolution of **4b**¹¹.

In order to further prove the stereopreference in the transesterifications, we subjected the remaining ester [(*R*)-**1c**] to enzyme catalysed methanolysis. The produced alcohol gave $[\alpha]_{20}^D = +21.66^\circ$, almost exactly opposite the reported values for the (*S*)-alcohol.

Microbial asymmetric reduction

The ketones **1a-4a** were subject either to fermenting cells or a cell free acetone powder¹² of *Geotrichum candidum* (IFO 4597). Only **1a** was reduced to give (*S*)-**1b**. This is in agreement with earlier reported substrate requirements for the oxido-reductases of *Geotrichum candidum*.¹² Fermenting cells gave an ee of 74 %, but high ee's (> 99 %) were obtained using acetone powder in buffer. However, the yields were not optimal, being 54 % in MES buffer and 78 % in phosphate buffer after 24 h. We tried to improve this by performing the reduction in organic solvents using *G. candidum* cells immobilized on Celite in hexane. Unfortunately this failed to give any measurable reduced product for any of the substrates **1a-4a**. A different system consisting of fermenting cells of *G. candidum*, immobilised on the water-absorbing polymer LP-100, also failed to give reductions for the substrates **2a-4a**. However, **1a** was reduced in 7 % yield after 4 days. This was considered too low to be of practical importance and further attempts were not carried out.

Experimental Section

General Procedures. Immobilized CALB (Novozyme 435, Novozymes) had an activity of 7000 PLU/g, and a water content of 1-2 % w/w. Solvents were dried over molecular sieves. The cultivation of *Geotrichum candidum* (IFO 4597) and preparations of its acetone-powder (AGP) are described elsewhere.¹² Column chromatography was performed using silica gel 60 from Fluka. Enzymatic reactions were performed in a shaker incubator (New Brunswick, Edison, NJ, USA). 1-(2-Thienyl)ethan-1-one (**1a**) was purchased from Fluka and the water absorbing polymer LP-100 *via* Sigma, Norway.

Analyses

Optical rotations were determined using an Optical Activity Ltd. AA-10 automatic polarimeter, concentrations are given in g/100 mL. NMR spectra were recorded in CDCl₃ solutions, using Bruker DPX 300 and 400 instruments, operating at 300 and 400 MHz for ¹H and 75 and 100 MHz for ¹³C, respectively. Chemical shifts are in ppm relative to TMS and coupling constants in Hz. Enantiomeric ratios, E were calculated on the basis of measurement of enantiomeric excess of both substrate (e_s) and product (e_p) at several degrees of conversion, using the computer program *E & K calculator version 2.03*¹³. Mass spectra were recorded on a Fison 8065/Fison TRIO 1000 GC-MS (EI, 70eV) system with a quadrupole mass filter, equipped with a DB-1701 column from J&W Scientific (25 m, 0.25 mm ID, 0.25 μm film thickness). Helium was used as carrier gas. Chiral analyses were performed using a Varian 3400 or 3800 gas chromatograph from Varian Instrument Group, Walnut Creek, California, USA. The columns were either Chiraldex G-TA, 10 m, 0.25 mm ID, 0.25 μm film thickness from Astec, Whippany, N.J., USA or CP-Chirasil-Dex CB, 25 m, 0.25 mm ID, 0.25 μm film thickness from Chrompack, Chrompack Norge A.S., Sandvika, Norway. The gas chromatographs were pressure regulated, carrier gas was hydrogen (Hydrogen 5.0, purity: 99.999%) with an outlet pressure of 3 bar (7 psi). Injection was performed in split mode at 200°C, split ratios 60. Detection was done using FID detectors at 200°C with air (300 mL/min) and hydrogen (30 mL/min) as flame gases. Integration was performed with Varian Star 4.0/4.5. The chromatographic details are given in Table 2.

Table 2. Columns, temperature programs and chromatographic properties of alcohols **1b–4b** and butanoates **1c–4c**

	Column	Temp. prog. Temp. increase 15°C/min	t _s	t _R	R _S
1b	Chirasil-dex	110 °C (11 min), 130 °C	11.84	12.34	2.55
1c	Chirasil-dex	110 °C (10 min), 180 °C	14.91	14.68	2.16
2b	Chiraldex GT-A	90 °C (10 min), 150 °C	8.60	9.22	3.10
2c	Chiraldex GT-A	130 °C (10 min), 180 °C#	12.42	12.62	1.34
3b	Chirasil-dex	130 °C (10 min), 180 °C	9.39	9.73	1.90
3c	Chiraldex GT-A	90 °C (10 min), 150 °C	7.64	8.04	1.86
4b	Chiraldex GT-A	80 °C (20 min), 150 °C#	22.12	23.34	1.35
4c	Chiraldex GT-A	85 °C (30 min), 150 °C#	32.54	34.02	1.23

#Much effort was put into getting baseline separation. This was not achieved in these cases.

Immobilisation and enzymatic reduction in hexane

AGP-celite

Celite was prepared washing with EtOH (2x) and then distilled water until the upper water was clear. AGP (100 mg) and NAD⁺ (20 mg) were suspended in MES buffer (2-(*N*-morpholino)ethanesulphonic acid-NaOH) (0.1M, pH 7.0, 3.0 mL) and left for hydration for 15 min, then mixed with the celite. The preparation was dried under vacuum overnight. Each reaction was performed in hexane (4 mL) by adding the immobilized APG (0.5 g), 2-propanol (100 µL) and substrate (20 mg).

Fermenting cells-LP-100

Geotrichum candidum was grown in 1L standard medium and filtered after 36 h to give 25g of wet cells.¹² The cells were re-suspended in distilled water (100 mL) and the water-absorbing polymer (LP-100, 15 g) was added. Each reaction was performed in hexane (6 mL) by adding the immobilised cells (4 g) and substrate (20 mg).

Small scale transesterifications

Substrate alcohol (2.2×10^{-4} mole) was dissolved in solvent (3 mL), vinyl butanoate (5 equivalents) was added and the reaction was started by adding immobilised CALB (10 mg) to the reaction mixture at 30°C. Chiral GLC analysis gave the enantiomeric excess of substrate (ees) and product (eep) from which conversion, *c*, was calculated, $c = ees / (ees + eep)$. In control experiments without enzyme, no acylation was observed using vinyl butanoate as acyl donor.

Enzymatic reduction in water (small scale)

The ketones **1a–4a** (0.013 mmole), NAD⁺ (7 µmole) and cyclopentanol or 2-propanol (100 µL) were added to a suspension of AGP (20 mg) in MES buffer, (0.1M, pH 7.0, 3 mL) or phosphate buffer (0.1M, pH 7.0, 3 mL). The mixture was shaken at 180 rpm at 30 °C. Small aliquots were withdrawn and extracted with Et₂O for GLC analyses. When GLC showed maximum yield, the

biomass was filtered off and filtrate saturated with NaCl, before extracting with Et₂O. The combined extracts were dried with MgSO₄ and evaporated. In control experiments without cofactor, no reduction was observed.

Reduction by fermenting cells

Geotrichum candidum was grown for 24 hours in 250 mL standard medium as described elsewhere.¹² The substrates (100 mg) was added to 1 mL of ethanol, added to the cultures and incubation continued using the same conditions. Progress of the reactions was measured by TLC. Workup and analysis was equal to the reduction by AGP in water.

1-(2-Thienyl)ethan-1-one (1a). 1-(2-Thienyl)ethan-1-one (**1a**) was obtained from Fluka. ¹H NMR δ 2.55 (3H, s, COCH₃), 7.11 (1H, dd, ³J = 3.85 and 4.82 Hz., thienyl H4), 7.62 (d, 1H, ³J = 3.85 Hz thienyl H3) and 7.70 (d, 1H, ³J = 4.82 Hz, thienyl H5) ppm. ¹³C NMR δ 26.7 128.5 132.5 133.8 144.5 and 190.7 ppm. MS: 126 (M⁺), 111 (100%), 83, 69, 57, 45, 43.

1-(2-Thienyl)propan-1-one (2a). Dry thiophene (9.3 mL, 0.118 mole), propanoic chloride (10.3 mL, 0.118 mole) and dry benzene (121.8 mL) was mixed and cooled to 0 °C. SnCl₄ (13.96 mL, 0.118 mole) was added dropwise over a 2h period. The cooling was then removed and the stirring continued for another hour. To the reaction mixture was then added HCl (conc, 6 mL) and ice water (55 mL). The organic layer was separated, the water phase extracted with CH₂Cl₂, the combined organic phase washed with water, dried with MgSO₄ and evaporated. The product was purified by column chromatography (pentane : Et₂O, 3 : 1) giving **2a** in 91 % yield. ¹H NMR δ 1.23 (3H, t, ³J = 7.4 Hz, CH₂CH₃), 2.94 (2H, q, COCH₂), 7.11 (1H, dd, ³J = 3.85 and 5.00 Hz, thienyl H4), 7.60 (d, 1H, ³J = 5.00 Hz, thienyl H5) and 7.69 (d, 1H, ³J = 3.85 Hz, H3) ppm. ¹³C NMR δ 8.9, 32.9, 128.7, 131.9, 133.6, 144.5, and 194.2 ppm. MS: 140 (M⁺), 111 (100%), 83, 69, 57, 45. TLC; R_f = 0.37 (pentane : Et₂O, 3 : 1).

1-(2-Thienyl)butan-1-one (3a). 1-(2-Thienyl)butan-1-one (**3a**) was synthesised by the same method as **2a**. Yield: 84.7 %. ¹H NMR δ 1.00 (3H, t, ³J = 7.4 Hz, CH₂CH₃), 1.79 (2H, m, CH₂CH₃), 2.84 (2H, t, ³J = 7.4 Hz, COCH₂), 7.12 (dd, 1H, ³J = 3.8 and 4.9 Hz, thienyl H4), 7.61 (d, 1H, ³J = 3.8, thienyl H3) and 7.71 (d, 1H, and ³J = 4.9 Hz, thienyl H5) ppm. ¹³C NMR: 13.8, 18.2, 41.3, 128.3, 131.6, 133.3, 144.6 and 192.5 ppm. MS: 154 (M⁺), 126, 111 (100%), 83, 57, 45. TLC; R_f = 0.37 (pentane : Et₂O, 3 : 1).

1-(2-Thienyl)pentane-1-one (4a). 1-(2-Thienyl)pentane-1-one (**4a**) was synthesised by the same method as **2a**. Yield: 72.4 %. ¹H NMR δ 0.94 (3H, t, ³J = 7.4 Hz, CH₂CH₃), 1.40 (2H, m, CH₂CH₃), 1.72 (2H, m, CH₂CH₂CH₃) 2.88 (2H, t, ³J = 7.3 Hz, COCH₂), 7.10 (1H, dd, ³J = 3.8 and 4.9 Hz, thienyl H4), 7.58 (d, ³J = 3.8 Hz, 1H, thienyl H5) and 7.69 (d, ³J = 3.8 Hz, 1H thienyl H3) ppm. ¹³C NMR δ 13.9, 22.6, 26.9, 39.3, 128.0, 131.2, 132.9 144.5 and 193.5 ppm. MS: 168 (M⁺), 139, 126, 111 (100%), 83, 57, 41. TLC; R_f = 0.37 (pentane : Et₂O, 3 : 1).

The racemic secondary alcohols **1b-4b** were synthesised from the ketones **1a-4a** by NaBH₄-reduction under standard conditions, quenching the reaction with NH₄Cl¹⁴.

1-(2-Thienyl)ethanol (1b). Yield: 90.3 %. ¹H NMR δ (in accordance with previously reported spectra⁶) 1.57 (d, 3H, ³J = 6.2 Hz, CHCH₃), 5.11 (q, 1H, ³J = 6.2 Hz, CHCH₃), 6.93-7.0 (m, 2H, thienyl H3 and H5) and 7.20-7.25 (m, 1H, thienyl H4) ppm. ¹³C NMR δ 25.6, 66.1, 123.1, 124.3,

126.6 and 150.0 ppm. MS: 128 (M^+), 113, 110, 95, 85 (100%), 66, 58, 45. TLC; R_f = 0.19 (EtOAc : hexane, 1 : 5).

1-(2-Thienyl)propanol (2b). Yield: 75.5 %. 1H NMR δ (in accordance with previously reported spectra) 0.94, (t, 3H, 3J = 7.4 Hz, CH_2CH_3), 1.85, (m, 2H, CH_2CH_3), 4.80, (t, 1H, 3J = 6.6 Hz, $CHOH$), 6.97 (2 d, 2H, 3J = 2.9 and 3.3 Hz, thienyl H3 and H5), 7.21 (1H, dd, 3J 2.9 and 3.3 Hz, thienyl H4). ^{13}C NMR δ 10.1, 32.2, 71.6, 123.7, 124.4, 126.5 and 148.7 ppm. MS: 142 (M^+), 124, 113 (100%), 97, 85, 57, 51, 45. TLC; R_f = 0.23 (pentane : Et₂O, 3 : 1).

1-(2-Thienyl)butanol (3b). Yield: 83.6 %. 1H NMR δ 0.93 (t, 3H, 3J = 7.4 Hz, CH_2CH_3), 1.40 (m, 2H), 1.80 (m, 2H), 4.87 (t, 1H, 3J = 6.7 Hz, $CHOH$), 6.93 (2 d, 2H, thienyl H3 and H5), 7.17 (2 d, 3J = 3.8 and 3.9 Hz, thienyl H4) ppm. ^{13}C NMR δ 13.8, 19.0, 41.4, 70.0, 123.6, 124.4, 126.5, 149.0 ppm. MS: 156 (M^+), 138, 123, 113 (100%), 97, 85, 45. TLC; R_f = 0.32 (pentane : Et₂O, 3 : 1).

1-(2-Thienyl)pentanol (4b). Yield: 70.2 %. 1H NMR δ 0.89 (t, 3H, 3J = 7.0 Hz, CH_2CH_3), 1.25-1.40 (m, 4H), 1.80 (m, 2H), 4.85 (t, 1H, 3J = 6.6 Hz, $CHOH$), 6.92 (2 d, 2H), 7.20 (2 d, 3J = 3.4' and 3.5 Hz) ppm. ^{13}C NMR δ 14.0, 22.5, 28.0, 38.0, 70.2, 123.6, 124.3, 126.5, 148.7 ppm. MS: 170 (M^+), 152, 129, 113, 110, 97, 85, 79, 45. TLC; R_f = 0.34 (pentane : Et₂O, 4 : 1).

Gram-scale resolutions (1.5 - 3 g) were performed in the solvent that gave the best result (high E-value and short reaction times) in small-scale transesterifications.

(R)-1-(2-Thienyl)ethyl butanoate [(R)-1c]. Racemic 1-(2-thienyl)ethanol (**1b**) (1.5 g) was dissolved in hexane (50 mL), vinyl butanoate (5 equivalents) was added and the reaction was started by adding immobilised CALB (200 mg) at 30°C/200 rpm. The reaction was monitored by chiral GLC. After 2.5 h the reaction virtually stopped and the enzyme was filtered off. The enantiomeric excess of both the produced ester and the remaining alcohol was 99 % or better. The ester and the alcohol were separated by column chromatography (EtOAc : hexane; 1 : 5), yield 0.91 g (78 %). 1H NMR δ 0.95 (t, 3H, 3J = 7.3 Hz, CH_2CH_3), 1.64 (m, 5H), 2.31 (t, 2H, 3J = 6.9 Hz), 6.19 (q, 1H, 3J = 6.6 Hz), 6.97 (2d, 1H, 3J = 3.3 Hz and 5.1 Hz), 7.05 (d, 1H, 3J = 3.3 Hz), 7.27 (d, 1H, 3J = 5.1 Hz) ppm. ^{13}C NMR δ 14.1, 18.9, 22.5, 36.9, 67.8, 125.1, 125.5, 127.0, 145.2 and 173.2 ppm. MS: 198 (M^+), 128, 111, 110, 85, 77, 71, 45. $[\alpha]_D^{20}$ = +107.2° (c 3.63, $CHCl_3$), +86.8° (c 4.72, EtOH).

(S)-1-(2-Thienyl)ethanol [(S)-1b]. Physical properties as for (*rac*)-**1b**, yield 0.47 g (63 %), = -21.3° (c 1.88, $CHCl_3$), -14.4° (c 1.88, EtOH).

(R)-1-(2-Thienyl)propyl butanoate [(R)-2c]. Resolution as for **1c**. 1H NMR δ 0.94 (m, 6H), 1.66 (m, 2H), 1.95 (m, 2H), 2.31 (t, 3H, 3J = 6.9 Hz), 6.00 (t, 1H, 3J = 6.9 Hz, $CHOH$), 6.96 (2 d, 1H, 3J = 3.5 and 3.4 Hz), 7.04 (d, 1H, 3J = 3.3 Hz), 7.25 (d, 1H, 3J = 3.4 Hz) ppm. ^{13}C NMR δ 10.5, 14.1, 18.9, 30.0, 36.9, 72.7, 125.7, 126.1, 127.0, 144.1 and 173.4 ppm. MS: 212 (M^+), 183, 142, 125, 124, 123, 113, 97, 71, 43. $[\alpha]_D^{20}$ = +111.6° (c 5.09, $CHCl_3$)

(S)-1-(2-Thienyl)propanol [(S)-2b]. Physical properties as for (*rac*)-**2b**. $[\alpha]_D^{20}$ = -25.4° (c 1.26, $CHCl_3$).

(R)-1-(2-Thienyl)butyl butanoate [(R)-3c]. Resolution as for **1c**. 1H NMR δ 0.95 (m, 6H), 1.38 (m, 2H), 1.67 (m, 2H), 1.98 (m, 2H), 2.31 (t, 2H, 3J = 6.9 Hz), 6.09 (t, 1H, 3J = 6.9 Hz), 6.97

(2d, 1H, $^3J = 3.5$ and 5.0 Hz), 7.03 (d, 1H, $^3J = 3.5$ Hz), 7.27 (d, 1H, $^3J = 5.0$ Hz). ^{13}C NMR δ 14.1, 18.6, 19.0, 19.4, 36.2, 39.0, 71.2, 125.0, 125.8, 126.1, 144.4 and 173.4 ppm. MS: 226 (M^+), 183, 156, 138, 123, 110, 98, 71, 43. $[\alpha]_{\text{D}}^{20} = +66.0^\circ$ (c 5.00, CHCl_3).

Conversion of (*R*)-1-(2-Thienyl)butyl butanoate [(*R*)-3c] into (*R*)-4-octanol

(*R*)-1-(2-Thienyl)butyl butanoate [(*R*)-3c] was hydrolysed by K_2CO_3 in MeOH to give (*R*)-1-(2-thienyl)butanol [(*R*)-3b]. (*R*)-3b (100 mg) was dissolved in MeOH (10 mL) and a suspension of Raney-Ni in water (2mL) was added. H_2 was added *via* a balloon and stirring was continued over night. The solution was filtered and evaporated, the residue was mixed with NaHCO_3 solution (5%, 5 mL) and extracted with CH_2Cl_2 (3x5mL). The combined extracts were dried and evaporated to give (*R*)-4-octanol (80 mg, 96.3% yield). $[\alpha]_{\text{D}}^{20} = +0.25^\circ$ (c 1.60, EtOH).

(*S*)-1-(2-Thienyl)butanol [(*S*)-3b]. Physical properties as for (*rac*)-3b. $[\alpha]_{\text{D}}^{20} = -51.5^\circ$ (c 1.00, CHCl_3).

(*R*)-1-(2-Thienyl)pentyl butanoate [(*R*)-4c]. Resolution as for 1c. ^1H NMR δ 0.92 (m, 6H), 1.33 (m, 4H), 1.65 (m, 2H), 1.93 (m, 2H), 2.28 (t, 2H, $^3J = 6.9$ Hz), 6.04 (t, 1H, $^3J = 7.0$ Hz), 6.94 (2d, 1H, $^3J = 3.5$ and 5.2 Hz), 7.03 (d, 1H, $^3J = 3.5$ Hz), 7.25 (d, 1H, $^3J = 5.0$ Hz). ^{13}C NMR δ 13.4, 13.7, 18.2, 22.1, 27.4, 35.9, 36.2, 70.8, 124.8, 125.4, 126.2, 143.7, 172.7 ppm. MS: 240 (M^+), 183, 170, 152, 123, 110, 97, 71, 60, 43. $[\alpha]_{\text{D}}^{20} = +44.4^\circ$ (c 4.01, CHCl_3).

(*S*)-1-(2-Thienyl)pentanol [(*S*)-4b]. Physical properties as for (*rac*)-4b. $[\alpha]_{\text{D}}^{20} = -17.9^\circ$ (c 1.00, CHCl_3).

Methanolysis of *R*-(1c).

R-1-(2-Thienyl)ethanol [(*R*)-1b]. The enantiopure ester (*R*)-1c (390 mg, 1.97 mmol) was added to 15 mL of hexane. Methanol (2 mL) and immobilized CALB (30 mg) were added to the reaction mixture and the reaction was left on a shaking incubator at 30°C for 72h. When normal GLC showed complete reaction, the enzyme was removed by filtration and solvent and the formed methylbutanoate was removed by rotavapor. The enantiomeric excess of (*R*)-1b was controlled by chiral GLC and showed the same ee as the ester (>99) and the product used without further purification. Yield 238 mg, 94.3%. $[\alpha]_{\text{D}}^{20} = +21.66^\circ$ (c 3.37, CHCl_3).

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