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First example of hydrolytic kinetic resolution of acrylate of secondary alcohols by lipase Amano AK

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ABSTRACT

Lipase Amano AK is found to be extremely efficient catalyst for hydrolytic kinetic resolution of acrylates of secondary alcohols in aqueous phosphate buffer at pH 7.0. Both aliphatic and benzylic secondary alcohols show good to excellent *E* values.

1. Introduction

In recent years, enzymes [1] are finding huge applicability in the manufacture of a wide range products, *viz.* drugs, agro-pharmaceuticals, organic fine chemicals and plastics [2] because of their easy commercial availability and high catalytic efficiency. Among the enzymes, lipases are used very often in organic synthesis because of their excellent selectivities. They catalyze hydrolysis, esterification, *trans*-esterification including alcoholysis on a broad range of substrates due to their ability to change their conformation depending on the substrate structure (induced fit enzyme) [3]. Especially, lipases are suited for the kinetic resolution of secondary alcohols due to their exceptional stability and enantioselectivity in both water and organic solvents, besides being environmentally friendly.

The α,β-unsaturated lactone [4] is one of the most important functionalities present in a wide spectrum of naturally occurring compounds that display diverse pharmacological properties, such as antitumor, antimicrobial, and antifungal. The synthesis of α,β-unsaturated lactones can be achieved by ring closing metathesis (RCM) of acrylates of secondary alcohol with unactivated olefin terminal [5]. The stereoselectivity of the acrylate terminal can be achieved in two pathways, *viz.* by esterification of chiral secondary alcohol, derived from non-enzymatic stereoselective reactions, with α,β-unsaturated acid chlorides [6], or through enzymatic kinetic resolution of acrylates of secondary alcohols (Scheme 1).

While surfing the literature, we have observed that the enzymatic esterification of olefin-tethered secondary alcohols to synthesize enantiomerically pure α,β-unsaturated ester is finding scant attention, albeit being very promising. In spite of having tremendous potential, there are only a few reports [7] on synthesis of chiral α,β-unsaturated esters by enzymatic *trans*- esterification of racemic secondary alcohols with α,β-unsaturated vinyl esters. But, no report is available for enzymatic hydrolysis of α,β-unsaturated esters leading to chiral alcohol, unlike the enzymatic hydrolysis of the acetate of secondary alcohols. It is a fact that in most of the kinetically controlled enzymatic reactions, equilibrium shift towards the right with slightest change in the reaction conditions. Therefore, both enzymatic esterification and hydrolysis are important pathways to achieve enantiomeric alcohol with high ee-values. In order to develop an efficient method for the synthesis enantiomerically pure acrylates and secondary alcohol, we are reporting for the first time, the lipase Amano AK catalyzed enzymatic kinetic hydrolysis of acrylates of secondary alcohols to achieve good to excellent enantioselectivity.

2. Experimental

2.1. General

All reagents were commercially available and used without further purification. Most of the alcohols were synthesized by $NaBH_4$ reduction of coommercially available ketones purchased

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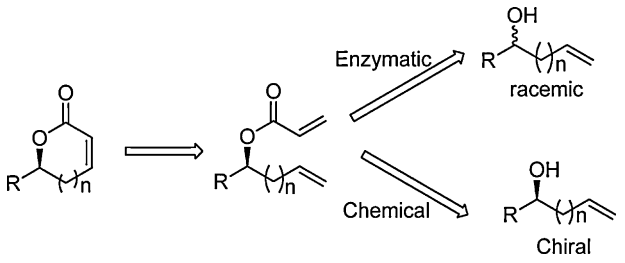


Scheme 1.

from Sigma Aldrich and acrylated with acryloyl chloride and DBU/triethylamine as per literature procedure [6a]. The alcohol derivatives for entry 6, 7 and 8 were synthesized by Grignard reaction with their corresponding aldehydes. All acrylates were characterized by ^{1}H NMR, ^{13}C NMR and IR spectroscopy. The IR spectra were recorded on a Perkin Elmer spectrophotometer. ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz) spectra were obtained on a Bruker AC-400 using $CDCl_3$ as solvent and TMS as internal standard, unless otherwise stated. Optical rotations were measured on a Perkin Elmer 341 polarimeter. HPLC analyses were performed on an Waters M515 series equipped with a chiral column (Chiralcel AD-H and Chiralcel OD-H), using mixtures of *n*-hexane/isopropyl alcohol (IPA) as mobile phase, at 25 °C. For column chromatography, we employed Merck silica gel 60–120 mesh.

2.2. General procedure for the synthesis of acrylates

A mixture of the secondary alcohol (2 mmol) and triethylamine (3 mmol) was dissolved in 20 mL dichloromethane and cooled to 0 °C. Then acryloyl chloride (5 mmol) was added drop wise to the reaction mixture and allowed to stir overnight. Upon completion, the reaction mixture was diluted with saturated $NaHCO_3$ solution and extracted with dichloromethane and aqueous NH_4Cl solution, dried over Na_2SO_4 and concentrated in a rotavapor. The residual oil is purified by column chromatography (silica gel, EtOAc/Hexane). The acrylates are characterized by ^{1}H NMR, ^{13}C NMR.

2.2.1. 1-Phenylethyl acrylate

Purification by column chromatography using hexane and ethyl acetate in 9:1 ratio to achieve 1-phenylethyl acrylate as a colorless liquid (yield 79%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.54 (d, J = 6.8 Hz, 3H), 5.78 (dd, J = 10.4 Hz, 1.6 Hz, 1H), 5.93 (q, J = 6.4 Hz, 1H), 6.11 (dd, J = 17.6 Hz, 10.4 Hz, 1H) 7.28 (m, 5H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 22.1, 72.7, 126.0, 127.9, 128.6, 130.7, 141.5, 165.4 ppm.

2.2.2. 1-(4-Methoxyphenyl)ethyl acrylate

Purification by column chromatography using hexane and ethyl acetate in 9:1 ratio to achieve 1-(4-methoxyphenyl)ethyl acrylate as a colorless liquid (yield 85%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.35 (d, J = 6.4 Hz, 3H), 3.58 (s, 3H), 5.59 (dd, J = 10, 1.2 Hz, 1H), 5.72 (q, J = 6.4 Hz, 1H), 5.91 (dd, J = 17.2, 10 Hz, 1H), 6.19 (dd, J = 17.6, 1.2 Hz, 1H), 6.70 (d, J = 8 Hz, 2H), 7.11 (d, J = 8.4 Hz, 2H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): δ 22.0, 55.3, 113.8, 127.6, 128.8, 130.7, 133.6, 165.5 ppm.

2.2.3. 1-(4-Chlorophenyl) ethyl acrylate

Purification by column chromatography using hexane and ethyl acetate in 9:1 ratio to achieve 1-(4-chlorophenyl)ethyl acrylate as a colorless liquid (yield 88%). ^{1}H NMR (400 MHz, $CDCl_3$): 0.55 (d, J = 6.8 Hz, 3H), 5.88 (dd, J = 27 Hz, 6.4 Hz, 1H), 5.84 (q, J = 6.4 Hz, 1H), 6.05 (dd, J = 17.2, 10.4 Hz, 1H), 6.34 (dd, J = 17.2, 0.8 Hz, 1H), 7.23 (m, 4H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 22.1, 71.7, 127.5, 128.4, 128.5, 128.7, 131.0, 133.6, 140.1, 165.3 ppm.

2.2.4. 1-(3-Nitrophenyl) ethyl acrylate

Purification by column chromatography using hexane and ethyl acetate in 9:1 ratio to achieve 1-(3-nitrophenyl)ethyl acrylate as a colorless liquid (yield 75%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.60 (d, J = 6.4 Hz, 3H) 5.88 (d, J = 10 Hz, 1H) 6.01 (q, J = 6.4 Hz, 1H), 6.15 (dd, J = 17.2 Hz, 10 Hz, 1H), 6.44 (d, J = 18 Hz, 1H), 7.52 (t, J = 8 Hz, 1H), 7.68 (d, J = 7.6 Hz, 1H), 8.14 (d, J = 8 Hz, 1H), 8.23 (s, 1H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 22.4, 71.4, 121.1, 123.0, 128.3, 129.7, 131.8, 132.4, 143.9, 165.3 ppm.

2.2.5. 1-(4-Methyl phenyl)ethyl acrylate

Purification by column chromatography using hexane and ethyl acetate in 9:1 ratio to achieve 1-(4-methylphenyl)ethyl acrylate as a colorless liquid (yield 82%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.52 (d, J = 6.8 Hz, 3H), 2.34 (s, 3H), 5.81 (dd, J = 10.4, 1.2 Hz,1H), 5.93 (q, J = 6.4 Hz, 1H), 6.17 (dd, J = 17.2, 10.4 Hz, 1H), 6.41 (dd, J = 17.6, 1.2 Hz, 1H), 7.164 (d, J = 8 Hz, 2H), 7.27 (d, J = 8 Hz, 2H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 21.1, 22.1, 72.4, 126.1, 128.8, 129.1, 130.7, 138.1, 165.5 ppm.

2.2.6. 1-Phenylbutyl acrylate

Purification by column chromatography using hexane and ethyl acetate in 12:1 ratio to achieve 1-phenylbutyl acrylate as a colorless liquid (yield 73%). ^{1}H NMR (400 MHz,$CDCl_3$): δ 0.92 (t, J = 14.8, 7.2 Hz, 3H), 1.31 (m, 2H), 1.86 (m, 2H), 5.81 (t, J = 2.4 Hz, 1H), 5.83 (d, J = 0.8 Hz, 1H), 6.15 (dd, J = 21.2, 10.4 Hz, 1H), 6.41 (d, J = 21.6 Hz, 1H), 7.30 (m, 5H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 13.8, 18.7, 38.4, 76.0, 126.4, 127.8, 128.4, 128.7, 130.7, 140.7, 165.5 ppm.

2.2.7. 1-(4-Bromophenyl)allyl acrylate

Purification by column chromatography using hexane and ethyl acetate in 12:1 ratio to achieve 1-(4-bromophenyl)allyl acrylate as a colorless liquid (yield 85%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 5.25 (t, J = 16.8 Hz, 2H), 5.83 (dd, J = 10.4, 1.2 Hz, 1H), 5.91–5.98 (m, 1H), 6.13 (dd, J = 17.2, 10.4 Hz, 1H), 6.24 (d, J = 5.6 Hz, 1H), 6.41 (dd, J = 17.2, 1.2 Hz, 1H), 7.21 (d, J = 8.4 Hz, 1H), 7.44 (d, J = 8.4 Hz, 1H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 75.6, 117.5, 122.2, 128.2, 128.9, 131.7, 135.6, 137.8, 165 ppm.

2.2.8. 1-[(E)-2-Phenylvinyl]but-3-enyl acrylate

Purification by column chromatography using hexane and ethyl acetate in 12:1 ratio to achieve 1-[(E)-2-phenylvinyl]but-3-enyl acrylate as a colorless liquid (yield 66%). ^{1}H NMR: δ 2.53 (m, 2H), 5.09–5.16 (m, 2H), 5.56 (q, J = 6.6 Hz, 1H), 5.75–5.85 (m, 2H), 6.11–6.21 (m, 2H), 6.43 (dd, J = 17.4, 1.4 Hz, 1H), 6.64 (d, J = 16.0 Hz, 1H), 7.24–7.39 (m, 5H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 39.1, 73.9, 118.2, 126.6, 126.9, 128.0, 128.5, 128, 6, 130.8, 132,7, 133.0, 136.2, 165.4 ppm.

2.2.9. Octan-2-yl acrylate

Purification by column chromatography using hexane and ethyl acetate in 20:1 ratio to achieve octan-2-yl acrylate as a colorless liquid (yield 86%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.79 (t, J = 6 Hz, 3H), 1.09–1.60 (m, 13H), 4.85–4.93 (m, 1H), 5.71 (dd, J = 10, 1.2 Hz, 1H), 6.02 (dd, J = 17.2, 10.4 Hz, 1H), 6.3 (d, J = 16.8 Hz, 1H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.0, 19.9, 22.5, 25.3, 29.1, 31.7, 35.8, 71.3, 129.1, 130.1, 165.9 ppm.

2.2.10. Octan-3-yl acrylate

Purification by column chromatography using hexane and ethyl acetate in 20:1 ratio to achieve octan-3-yl acrylate as a colorless liquid (yield 81%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.71–0.81 (m, 6H), 1.18 (m, 6H), 1.41–1.55 (m, 4H), 4.80 (m, 1H), 4.84 (dd, J = 10.4, 1.2 Hz, 1H), 6.02 (dd, J = 17.6, 10.4 Hz, 1H), 6.3 (dd, J = 17.6, 1.2 Hz,

1H) ppm; ^{13}C NMR (100 MHz, $CDCl_3$): δ 9.6, 14, 22.5, 24.9, 26.9, 31.5, 75.7, 129, 130.2, 166.1 ppm.

2.2.11. Hept-1-en-3-yl acrylate (3b)

Purification by column chromatography using hexane and ethyl acetate in 20:1 ratio to achieve hept-1-en-3-yl acrylate (3b) as a colorless liquid (yield 88%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.82 (t, J = 6.4 Hz, 3H), 1.18–1.3 (m, 4H), 1.52–1.63 (m, 2H), 5.08–5.26 (m, 3H), 5.69–5.78 (m, 2H), 6.06 (dd, J = 17.2, 10.4 Hz, 1H), 6.34 (dd, J = 17.6, 1.6 Hz, 1H) ppm. ^{13}C NMR δ (100 MHz, $CDCl_3$): 13.9, 22.4, 27.2, 33.9, 75, 116.6, 128.8, 130.5, 136.5, 165.5 ppm.

2.2.12. (R)-5-Butyl-2(5H)-furanone 3f

Purification by column chromatography using hexane and ethyl acetate in 12:1 ratio to achieve (*R*)-5-butyl-2(5*H*)-furanone (3f) as a colorless liquid (yield 91%). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.84 (t, J = 6.8 Hz, 3H), 1.25–1.4 (m, 4H), 1.55–1.74 (m, 2H), 4.96–5 (m, 1H), 6.03 (dd, J = 5.6, 2 Hz, 1H), 7.42 (dd, J = 5.6, 1.2 Hz, 1H) ppm. ^{13}C NMR (100 MHz, $CDCl_3$): δ 13.8, 22.4, 27, 83.5, 121.4, 156.7, 173.3 ppm.

2.3. Preparation of phosphate buffer

One Buffer Tablet pH 7.0 containing chlorides, potassium phosphates and sodium phosphates, purchased from Fluka (Code no. 82561-1EA), was dissolved in milipore water in a 100 mL volumetric flask and made up to the 100 mL mark to prepare pH 7.0 phosphate buffer solution. The pH of the solution was ascertained by digital pH meter (DP.505) and used for hydrolysis of acrylates in the presence of lipases.

2.4. General procedure for lipase Amano AK catalyze hydrolysis of acrylates

The racemic acrylates (0.1 g) was taken in 20 mL aqueous phosphate buffer and to it 0.05 g lipase Amano AK, from *Pseudomonas fluorescens*; (Purchased from Sigma–Aldrich: CAS 9001-62-1; Item code: 534730-50G; Batch # 07919JE; EC 232-619-9, WGK1) was added. The mixture was stirred at room temperature and the progress of the reaction was monitored from time to time by TLC. The pH of the reaction mixture was monitored and no substantial change in pH was observed due to formation of the acrylic acid byproduct during the course of reaction. After almost 50% conversion of the starting material (from TLC), the reaction mixture was extracted with ethyl acetate (3× 10 mL), dried over Na_2SO_4 and concentrated under vacuo. The resulting mixture was separated by column chromatography using mixture ethyl acetate and hexane.

2.4.1. Specific rotation and HPLC data

(R)-PHENYLETHAN-1-OL: 90% ee. $[\alpha]^D_{20}$ = +38.5 (c 1.0, $CHCl_3$) [lit. [8]; $[\alpha]^D_{20}$ = +48.6 (c 1.0, CH_2Cl_2)]. HPLC analysis: Chiralcel OD-H, *n*-hexane/2-propanol (90:10), 0.5 mL/min, 217 nm; t_R: (*R*) 11.08 min, (*S*) 12.50 min.

(R)-1-(4-METHOXYPHENYL)ETHAN-1-OL: 98% ee. $[\alpha]^D_{20}$ = +48.5 (c 1.0, $CHCl_3$) [lit. [8]; $[\alpha]^D_{20}$ = +47.2 (c 1.0, $CHCl_3$)]. HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (95:5), 0.5 mL/min, 217 nm; t_R: (*R*) 21.97 min, (*S*) 32.34 min.

(R)-(4-CHOLROPHENY)LETHAN-1-OL: 87% ee. $[\alpha]^D_{20}$ = +47.1 (c 1.0, $CHCl_3$) [lit. [8]; $[\alpha]^D_{20}$ = +46.1 (c 1.0, Et_2O)]. HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (90:10), 0.5 mL/min, 220 nm; t_R: (*R*) 28.79 min; (*S*) 40.89 min.

(+)-1-(3-NITROPHENYL)ETHAN-1-OL: 30% ee. $[\alpha]^D_{20}$ = +54.5 (c 1.0, $CHCl_3$). HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (95:5), 0.4 mL/min, 220 nm; t_R:(*R*) 16.97 min; (*S*) 17.62 min.

(R)-1-(4-METHYLPHENYL)ETHAN-1-OL: 96% ee. $[\alpha]^D_{20}$ = +39.5 (c 1.0, $CHCl_3$) [lit. [8]; $[\alpha]^D_{20}$ = +51.1 (c 1.0, $CHCl_3$)]. HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (95:5), 0.5 mL/min, 220 nm; t_R: (*R*) 14.53 min, (*S*) 16.38 min.

(R)-1-PHENYLBUTAN-1-OL: 82% ee. $[\alpha]^D_{20}$ = +59 (c 1.0, $CHCl_3$). HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (90:10), 0.5 mL/min, 220 nm; t_R: (*R*) 10.94 min, (*S*) 12.04 min.

(R)-1-(4-BROMOPHENYL)PROP-2-EN-1-OL: 94% ee. $[\alpha]^D_{20}$ = +46.4 (c 1.0, $CHCl_3$) HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (95:5), 0.5 mL/min, 254 nm; t_R: (*S*) 10.15 min, (*R*) 10.72 min.

(3R)-(1E)-PHENYLHEXA-1,5-DIEN-3-OL: 91% ee. $[\alpha]^D_{20}$ = +33.4 (c 1.0, $CHCl_3$). HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (95:5), 0.4 mL/min, 234 nm; t_R: (*R*) 28.29 min, (*S*) 31.08 min.

(S)-OCTAN-2-OL: $[\alpha]^D_{20}$ = +7.0 (c 1.0, $CHCl_3$) [9].

(S)-OCTAN-3-OL: $[\alpha]^D_{20}$ = +6.1 (c = 1, $CHCl_3$) [lit. [10]; $[\alpha]^D_{20}$ = +9.15 (c 4.33, $CHCl_3$)].

(S)-1-PHENYL ETHYL ACRYLATE: 87% ee. $[\alpha]^D_{20}$ = −48.6 (c 1.0, $CHCl_3$) HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (95:5), 0.5 mL/min, 254 nm; t_R: (*R*) 9.72 min, (*S*) 9.25 min.

(S)-1-(4-METHOXYPHENYL) ETHYL ACRYLATE: 99% ee. $[\alpha]^D_{20}$ = −16.4 (c 0.5, $CHCl_3$). Determination of the ee by HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (98:2), 0.5 mL/min, 254 nm; t_R (*R*) 12.98 min, (*S*) 17.89 min.

(S)-1-(4-CHLOROPHENYL) ETHYL ACRYLATE: 79% ee. $[\alpha]^D_{20}$ = −68 (c 1.0, $CHCl_3$). HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (98:2), 0.5 mL/min, 254 nm; t_R: (*R*) 11.56 min, (*S*) 9.76 min.

(-)-1-(3-NITROPHENYL) ETHYL ACRYLATE: 40% ee. $[\alpha]^D_{20}$ = −36.5 (c 1.0, $CHCl_3$). HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (98:2), 0.5 mL/min, 254 nm; t_R: (*R*) 16.34 min, (*S*) 19.99 min.

(S)-1-(4-METHYLPHENYL) ETHYL ACRYLATE: 63% ee. $[\alpha]^D_{20}$ = −86 (c 1.0, $CHCl_3$). HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (95:5), 0.5 mL/min, 254 nm; t_R: (*R*) 8.53 min, (*S*) 9.45 min.

(S)-1-PHENYL BUTYL ACRYLATE: 15% ee. $[\alpha]^D_{20}$ = −18.5 (c 1.0, $CHCl_3$). HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (98:2), 0.5 mL/min, 254 nm; t_R: (*R*) 7.89 min, (*S*) 9.24 min.

(1S)-1-(4-BROMOPHENYL)ALLYL ACRYLATE: 98% ee. $[\alpha]^D_{20}$ = −45.0 (c 1.0, $CHCl_3$). HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (98:2), 0.5 mL/min, 254 nm; t_R: (*R*) 20.93 min, (*S*) 32.70 min.

(1S)-1-[(E)-2-PHENYLVINYL]BUT-3-ENYL ACRYLATE: 92% ee. $[\alpha]^D_{20}$ = −48.6 (c 1.0, $CHCl_3$) [lit. [11]; $[\alpha]^D_{20}$ = −50.6 (c 2.1, $CHCl_3$)]. HPLC analysis: Chiralcel AD-H, *n*-hexane/2-propanol (98:2), 0.5 mL/min, 254 nm; t_R: (*R*) 10.35 min, (*S*) 11.47 min.

(R)-2-OCTANYL ACRYLATE: $[\alpha]^D_{20}$ = −18 (c 1.0, $CHCl_3$).

(R)–3-OCTANYL ACRYLATE: $[\alpha]^D_{20}$ = −14 (c 1.0, $CHCl_3$).

2.5. Preparation of hept-1-en-3-yl acrylate, 3e

To an ice cold solution of (*R*)-3-hydroxyhept-1-ene, 3d (0.1 g, 0.88 mmol) and DBU (0.2 mL, 1.3 mmol) in dichloromethane (15 mL), acryloyl chloride (0.17 mL, 2.2 mmol) was added dropwise at 0 °C and allowed to stir for 12 h. Upon completion, the reaction mixture was diluted with saturated aqueous $NaHCO_3$ solution and extracted with dichloromethane (30 mL ×3). The organic layer was dried over anhydrous Na_2SO_4 and concentrated to get the crude product, which was purified by column chromatography using 5% ethyl acetate in hexane to achieve the product (0.143 g, 99%) as a colorless liquid.

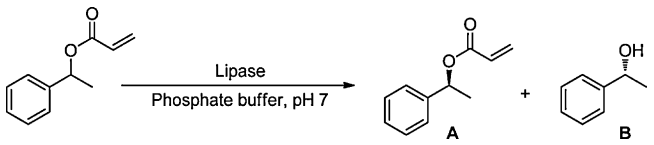
2.6. Synthesis of (R)-5-Butyl-2(5H)-furanone, 3f

To a solution of hept-1-en-3-yl acrylate, 3e (0.075 g, 0.45 mmol) in dry dichloromethane (10 mL), Grubb's catalyst 2nd generation (0.019 g, 5 mol%) was added and allowed to stir at room temperature under nitrogen for 12 h. After completion of the reaction, the solvent was removed under vacuum and the crude product was immediately charged into silica gel column and eluted with ethyl

acetate and hexane mixture in 12:1 ratio to achieve the desired product, 3f (0.057 g, 91%) as a colorless liquid.

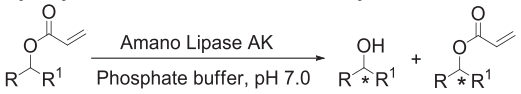
3. Results and discussion

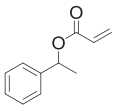
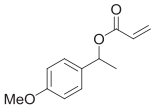
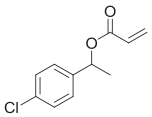
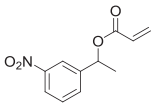
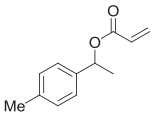
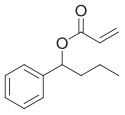
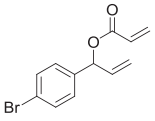
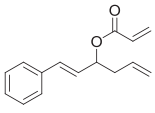
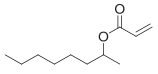
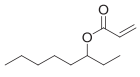
Since lipase Amano AK is finding a variety of applications in organic synthesis, such as stereoselective *trans*-esterification for resolution of alcohols and alcoholysis [12], we decided to carry out enzymatic hydrolysis of (±)-1-phenyl ethyl acrylate in the presence of Lipase Amano AK at pH 7.0 in aqueous medium (Scheme 2). Although, the acrylate did not dissolve in water, we were excited to see that enzymatic hydrolysis does take place under this condition. The reaction was monitored by TLC and stopped after 30 h, when approximately 50% consumption of the starting material was observed. The reaction mixture was extracted with ethyl acetate (15 mL ×3), dried (over Na_2SO_4), concentrated under vacuum and separated by column chromatography. HPLC analysis using Chiralcel AD-H column with 10% isopropanol in hexane as eluent showed



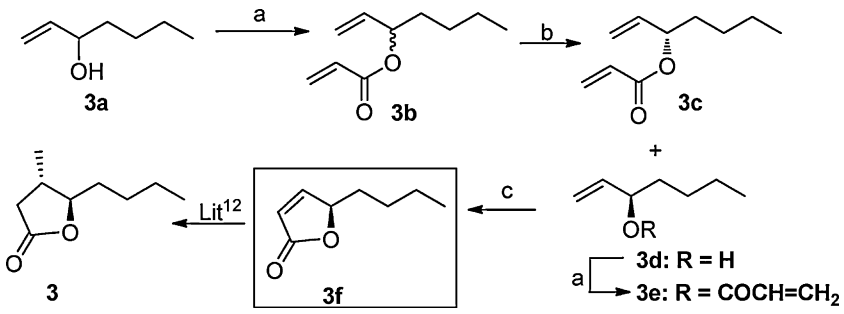
Scheme 2.

Table 1
Hydrolytic kinetic resolution of racemic acrylates



Entry	Substrate	Time (h) %	yield[a] (A/B)	% ee of A[b]	% ee of B[b]	*E*-value[c]	Configuration[d] (A/B)
1		30	38/42	90	87	52	R/S
2		32	47/36	98	99	>500	R/S
3		32	40/44	87	79	35	R/S
4		45	39/44	30	40	3	ND
5		34	44/40	96	63	94	R/S
6		62	31/47	82	15	12	R/S
7		6	45/46	94	98	>100	R/S
8		40	45/42	91	92	70	R/S
9		72	42/43	ND	ND	ND	S/R
10		80	42/45	ND	ND	ND	S/R

[a] Isolated yield, after purification by column chromatography.
[b] Enatiomeric excess was determined by HPLC with Chiralcel OD-H and AD-H columns.
[c] $E = \ln[(1 - eeS)/(1 + eeS/eeP)]/\ln[(1 + eeS)/(1 + eeS/eeP)]$.
[d] Absolute stereochemistry of the alcohols were determined by comparing the sign of optical rotation the alcohols in the literature [8–11]. ND = not determined.



Scheme 3. Reagent and conditions: (a) acrolyl chloride, DBU, 99%; (b) lipase Amano AK, phosphate buffer (pH 7.0), 3 days, 48%; (c) 2nd generation Grubb's catalyst, N_2 atmosphere, 12 h, 91%.

excellent enantiomeric excess (87% ee) for the acrylate and the alcohol (90% ee). Comparison of specific rotations of the both the acrylate and the alcohol with literature report [8] led us to conclude that the acrylate has *S*-configuration, while the alcohol has the opposite, i.e. the *R*-configuration.

Since the enzyme enantioselectivity, *E* for this particular conversion was moderate ($E = 52$) in comparison to hydrolysis of acetate ($E > 300$) of the same alcohol [13], we set out to screen other lipases at our disposal in order to compare their reactivity and enzyme selectivity with lipase Amano AK and study the effect of water–acetonitrile mixture (9:1) on the reaction profile. It was observed that for (±)-1-phenyl ethyl acrylate, the reaction in water-acetonitrile mixture (9:1) solvent gave higher yield (43%) of the hydrolyzed product, i.e. 1-phenethyl alcohol. But enantioselectivity of the alcohol got reduced substantially in water–acetonitrile medium (10% ee) in comparison to water medium (Table 1). In the hydrolysis of phenylethyl acrylate in water medium using lipase *Amano PS*, 50% conversion of the starting material was not achieved even after 120 h and gave only 25% isolated yield with 20% ee. In the cases of *Candida rugosa lipase* and *Aspergillus niger lipase*, continuous stirring of the reaction mixture for 144 h were not enough for 50% conversion of the starting material under similar conditions and the selectivities of the isolated alcohol were not appreciable either, 5% ee and 18% ee respectively. In water–acetonitrile medium, no appreciable incremental changes in reactivity and selectivity were observed for all the aforesaid lipases in the hydrolysis of (±)-1-phenyl ethyl acrylate. When the same reaction was carried out with the lipases, such as *Mucor javanicus lipase* (entry 4) and PPL (entry 6), they were found not selective under our reaction conditions.

As lipase is known to sustain temperature as high as 70 °C, we sought to study the effect of temperature on selectivity of Amano AK under hydrolytic conditions. Ironically, increase of temperature showed adverse effect on enantioselectivity. When temperature was enhanced to 30 °C, 50% conversion of (±)-1-phenyl ethyl acrylate took almost similar time, but the enantioselectivity of the hydrolyzed product dropped down to 57% ee. At 35 °C, the reaction time could be reduced to 16 h for 50% conversion, but ended up with only 19% ee of the resulting alcohol. Further increase of temperature to 40 °C resulted in no selectivity, although it took only 10 h for 50% conversion of ester.

Having standardize all the reaction parameters, we explored the reactivity and selectivity pattern in other benzylic alcohols (Table 1, entry 2–6)) and they were found to have reasonably high enantioselectivity except 1-(3-nitrophenyl) ethanol (entry 4), in which case poor enantioselectivity ($E = 3$) was observed. Nevertheless, the introduction of longer chain in the benzylic alcohol (entry 6, Table 1) might have affected the efficiency of the catalyst system as evident from the poor conversion to alcohol even after 62 h, albeit giving reasonably good %ee of the alcohol. Interestingly, the substrates having vinylic alcohol moiety react comparatively faster and showed good enantioselectivity as evident from entry 7 and 8 in Table 1. The acrylate of allylic secondary alcohol, 1-(4-bromophenyl)-prop-2-en-1-ol (entry 7, Table 1) underwent enzymatic hydrolysis in the presence of Lipase Amano AK at pH 7.0 in aqueous phosphate buffer to give excellent selectivity for both the alcohol (94% ee) and the acrylate (98% ee). It is interesting to note that when the acrylate is flanked by two π-systems (entry 8, Table 1), hydrolysis is very fast (6 h) under our reaction conditions and gave very good enantioselectivities for both the alcohol (91% ee) and the acrylate (92% ee).

We sought to examine lipase Amano AK catalyzed hydrolysis of acrylates for some saturated aliphatic alcohols to explore the versatility of this method. The acrylates of 2-octanol and 3-octanol were giving (*S*)-alcohol upon treatment with lipase under similar hydrolytic reaction conditions. The (*S*)-configuration of 2-octanol and 3-octanol were confirmed by comparimg the sign of specific rotation of reported in the literature [9,10], while the (*R*)-configuration of acrylates were assigned by comparing the sign of rotation of corresponding (*S*)-alcohols. Moreover, hydrolysis of these acrylates took comparatively longer time than benzylic alcohols, which might be due to greater hydrophobicity of those esters that comparatively hinders their entry to the enzyme reaction site. Here too, the acrylate of octan-2-ol (entry 9, Table 1) reacted faster (72 h for approximately 50% conversion) that the acrylate of octan-3-ol (entry 10). For the acrylate of allylic secondary alcohol, 3b we observed the preference for hydrolysis of (*R*)-acrylate in spite having aliphatic chain (Scheme 3). The configuration of the alcohol, 3d was confirmed by synthesizing (*R*)-5-Butyl-2(*5H*)-furanone 3f, the penultimate precursor to (*R*)-(+)-*trans*-Whisky lactone, 3 (Scheme 3) and comparing the sign of specific rotation. The synthesis started with acrylation of the allylic alcohol, 3a with acryloyl chloride and DBU to achieve the acrylate, 3b followed by hydrolytic kinetic resolution in aqueous medium at pH 7.0 (using phosphate buffer) to get 3c (48% yield; $[\alpha]^{D}_{20} = +9.8$ (c= 1.5, $CHCl_3$) and 3d (45% yield). Acrylation of 3d followed by RCM reaction using Grubb's 2nd generation catalyst led to 3f {$[\alpha]^{D}_{20} = -101.0$ (c = 2.0, $CHCl_3$); lit. [11]; $[\alpha]^{D}_{20} = -112.0$ (c 1.57, MeOH)}, the penultimate precursor of (*R*)-(+)-*trans*-whisky lactone.

4. Conclusion

In conclusion, we have for the first time reported that acrylates of secondary alcohols efficiently undergoes hydrolytic kinetic resolution in the presence of lipase Amano AK, in that case, in the presence of lipase to generate enantiomerically pure secondary alcohols and their acrylates with very good *E*-values. The method works extremely well for benzylic and other secondary alcohols. The method may find useful applications for the synthesis of α,β-unsaturated lactones.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.molcatb.2011.06.015.

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