SUPPORTING INFORMATION

Enantioselective Aminolysis of α-Chloroester Catalyzed by *Candida cylindracea* Lipase Encapsulated in Sol-Gel Glass

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1. Instrumentation

Ultraviolet-visible (UV-vis) spectra were recorded with a Perkin Elmer Lambda 18 spectrophotometer. Nuclear magnetic resonance (NMR) spectra were recorded with a Bruker DRX-400 spectrometer; integration error was 5–10 %. Gas chromatography–mass spectrometry (GC-MS) experiments were done with a Varian gas chromatograph attached to a Finnigan TSQ 700 mass spectrometer configured in the electron-impact ionization mode. The first quadrupole scanned the m/z values from 35 to 650, at a rate of 1.2 scans per second. The second and third quadrupoles were kept in the RF-only mode. Unit mass resolution was achieved using FC43 as the calibration and tuning reference. The amide enantiomers were separated isothermally with a Chiraldex B-DM GC column sized 30 m x 0.25 mm as follows: at 160 °C for *N*-*p*-methoxyphenyl-2-chloropropionamide, at 145 °C for *N*-*p*-tolyl-2-chloropropionamide and *N*-benzyl-2-chloropropionamide, at 112 °C for *N*-cyclohexyl-2-chloropropionamide, at 90 °C for *N*-*n*-butyl-2-chloropropionamide, and at 70 °C for *N*-*tert*-butyl-2-chloropropionamide; integration error was 3–5 %.

$$ee(A) = ee(E)\frac{c}{1-c} \tag{S1}$$

This column could not separate enantiomers of *N*-allyl-2-chloropropionamide. The enantiomeric excess for this amide, designated *ee(A)*, was estimated from that of the starting ethyl 2-chloropropionate, designated *ee(E)*, according to eq. S1, in which *c* is the conversion of racemic ester.[1] In the GC separation of the ester enantiomers, the temperature was kept at 70 °C for 1 min and than raised to 100 °C at a rate of 4 °C per minute.

2. Chemicals

Candida cylindracea lipase (designated *Cc*L), *p*-anisidine, *p*-toluidine, and *tert*-butylamine were obtained from Fluka Chemical Co. Ethyl 2-chloropropionate, 2-chloropropionyl chloride, allylamine, *n*-butylamine, cyclohexylamine, tetramethylorthosilicate (tetramethoxysilane), trimethoxypropylsilane, poly(vinyl alcohol), and triphenylphosphine were obtained from Aldrich Chemical Co. Hydrochloric acid was obtained from Fisher

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[1] Chen. C.-S.; Fujimoto, Y.; Girdaukas, G.; Sih, C. J. *J. Am. Chem. Soc.* **1982**, *104*, 7294-7299.

Scientific Co. Benzylamine was obtained from Acros Chemical Co. Merrifield resin (a copolymer of styrene and 1 % divinylbenzene containing benzyl chloride groups) sized 100–200 mesh and having a loading of 1.46 mmol chlorine per g was obtained from Novabiochem Co. Coomassie Blue for protein assays was obtained from Bio-Rad Co. as a "protein assay dye reagent concentrate" and used according to the manual. Dichloromethane-d_2 was obtained from Cambridge Isotope Laboratories, Inc. Distilled water was demineralized to electrical resistivity greater than 17 MΩ•cm.

3. Encapsulation of lipase into hydrophobic sol-gel glass

A suspension of 20.0 g of crude *C. cylindracea* lipase in 80.0 mL of deionized water was vigorously stirred for 2 min and shaken for 1 h. After removal of insoluble material by centrifugation overnight, the solution was lyophilized to dryness at –70 °C for 24 hours. The resulting beige powder (*ca.* 9.3 g), which is the water-soluble fraction of the crude lipase, was combined with water (23.24 mL), aqueous sodium fluoride (1.00 M, 5.57 mL), and aqueous poly(vinyl alcohol) (4 % w/w, 11.14 mL). After the solid dissolved, the solution in several plastic centrifuge tubes was vigorously stirred in a vortex shaker while adding trimethoxypropylsilane (total volume 48.75 mL) and then tetramethylorthosilicate (total volume 8.245 mL), to obtain a clear solution. During stirring for additional 5 min gelation occurred. The sealed centrifuge tubes were kept at room temperature for 24 h and then opened. The gel was air-dried at room temperature for 5 days. After grinding in a mortar, the solid was washed with water, acetone, and hexane; and air-dried for 2 days. The final product was *ca*. 25 g of an off-white powder.

4. General procedure for aminolysis reaction

Two forms of the enzyme were used in the reaction: 0.900 g of lipase encapsulated into hydrophobic sol-gel glass, designated *Cc*L@glass; and 318.4 mg of water-soluble fraction of lipase, designated ws*Cc*L. In a typical experiment, catalyst was suspended in 5 mL of CCl_4. Solutions of ester (1.000 mmol) and amine (0.500 mmol), each in 2 mL of CCl_4, were added to the catalyst suspension. Reaction was followed by ^{1}H NMR and chiral GC-MS methods. The compounds were identified by their GC retention times, mass spectra, and ^{1}H NMR chemical shifts, in comparison with authentic samples and published data.[2]

In the recycling experiments, the catalyst was filtered off after each cycle, thoroughly washed with CCl_4 and acetone, and air-dried for 10 min. The racemizing agent, triphenylphosphonium chloride bound to Merrifield resin, was prepared by a published procedure.[3] In the experiments concerning dynamic kinetic resolution, 1.000 mmol of racemic amine and 1.000 mmol of this agent were used.

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[2] Gotor, V.; Brieva, R.; Gonzalez, C.; Rebolledo, F. *Tetrahedron* **1991**, *47*, 9207-14.

[3] Zheng, A.; Shan, D.; Shi, X.; Wang, B. *J. Org. Chem.* **1999**, *64*, 7459-7466.