Novel Plots of Data from Combined Multistep Enzymatic Resolutions of Enantiomers

Zhi-Wei Guo

School of Pharmacy, West China University of Medical Sciences, Chengdu, Sichuan 610044, The People's Republic of China

Received March 12, 1993 (Revised Manuscript Received July 19, 1993•)

Novel plots of data from combined multistep enzymatic resolutions of enantiomers are disclosed and used to predict the maximal chemical recovery of the final product with a fixed enantiomeric excess and the corresponding conversions of all steps. The predicted values are verified experimentally. Through two porcine pancreatic lipase-catalyzed enantioselective esterifications of (±)-2-(6-methoxy-2-naphthyl)propanol (1) with vinyl acetate and product recycling, (*S*)-1 (naproxol) is prepared in high chemical and optical yields.

Introduction

Enzymatic resolution has been extensively used as an alternative method for the preparation of optically active substances.[1] For many unnatural substrates, catalysis often occurs with low to moderate enantioselectivity. In these cases, strategies such as product recycling,[2] coupled enantioselective action,[3] and sequential biocatalytic resolution[4] have been used to enhance the enantiomeric excess (ee) of the product of the enzymatic reaction.

Quantitative analyses based on eq 1 for irreversible enzymatic reaction steps, including cases when ee0 = 0 and when ee0 > 0, have been given in detail by Sih,[2] where

$$1 - C[(1 + eep)/(1 + ee0)] = [1 - C[(1 - eep)/(1 - ee0)]]^{E} \quad (1)$$

In a product recycling case, the product pair of enantiomers of the first step (PP1) is nonselectively transformed into the starting substrate pair of enantiomers of the second step (SP2), and the resolutions of both steps are the same. The key point is that the ee of SP2 is equal to the ee of PP1, and one symbol (ee1) is employed for both, showing the correlation between the two combined resolution steps. Generally, combined multistep resolutions consist of several (n) enantioselective steps with the same enantiomeric preference, in which the ee of the PP of any step is equal to the ee of the SP of the next step (Scheme I).

Scheme I

$$\underset{(ee_{i-1})}{SPi} \xrightarrow{E_i} \underset{(eei)}{PPi} + \underset{(eesi)}{RPi} \quad (i = 1, 2, \cdots, n)$$

In Scheme I, i is the serial number of the step, E_i is the enantioselectivity of step i, ee_{i-1} and eei denote the ee of the fast-reacting enantiomer of SPi and PPi, respectively, and eesi is the ee of the slow-reacting enantiomer of RPi (the pair of substrate enantiomers that remains after step i). Substituting these values into basic eq 1 gives eq 2 for step i where Ci is the percent conversion of step i. In this

$$1 - Ci[(1 + eei)/(1 + ee_{i-1})] = [1 - Ci[(1 - eei)/(1 - ee_{i-1})]]^{E_i} (i = 1, 2, \cdots, n) \quad (2)$$

$$Ci = (eesi + ee_{i-1})/(eesi + eei) \quad (3)$$

work, the theoretical chemical recovery (TCR) of the final product with a fixed ee$n$ for the combined n-step resolution system is based on conversions of all steps in the process.

Results and Discussion

Computer Programs EEP and C. When E_i and ee_{i-1} were known for any single step i, eq 2 as an implicit function was solved by iteration[5] based on eq 4 (program EEP).

$$\begin{cases} eei = 1 - [1 - [1 - Ci[(1 + eei)/(1 + ee_{i-1})]]^{1/E_i}](1 - ee_{i-1})/Ci & (4a) \\ \text{or} \\ eei = -1 + [1 - [1 - Ci[(1 - eei)/(1 - ee_{i-1})]]^{E_i}](1 + ee_{i-1})/Ci & (4b) \end{cases}$$

The output data with program EEP made eei a numerical function of the variable Ci when E_i and ee_{i-1} were known. Similarly, the output data with program C based on eq 5 made Ci a numerical function of ee_{i-1} when E_i was known and eei was fixed.

$$\begin{cases} Ci = [1 - [1 - Ci[(1 + eei)/(1 + ee_{i-1})]]^{1/E_i}](1 - ee_{i-1})/(1 - eei) & (5a) \\ \text{or} \\ Ci = [1 - [1 - Ci[(1 - eei)/(1 - ee_{i-1})]]^{E_i}](1 + ee_{i-1})/(1 + eei) & (5b) \end{cases}$$

Computer Program C2SR. Program C2SR was compiled for quantitative analyses of combined two-step resolutions. E_1, E_2, and ee0 were determined, and ee2 was fixed at an appropriate value. Then C2 was made a numerical function of C1 by using program EEP for the first step (i = 1) and program C for the second (i = 2). Furthermore, the TCR for the whole process, equal to C1C2, was also made a numerical function of C1, and the maximal chemical recovery (MCR) and the corresponding C1 and C2 were determined by the program. The useful theoretical curves were plotted.

• Abstract published in *Advance ACS Abstracts*, September 1, 1993.

(1) (a) Sih, C. J.; Wu, S. H. *Top. Stereochem.* **1989**, *19*, 63–70. (b) Chen, C. S.; Sih, C. J. *Angew. Chem., Int. Ed. Engl.* **1989**, *28*, 695–707. (c) Wong, C. H. *Science* **1989**, *244*, 1145–1152. (d) Klibanov, A. M. *Acc. Chem. Res.* **1990**, *23*, 114–120.

(2) Chen, C. S.; Fujimoto, Y.; Girdaukas, G.; Sih, C. J. *J. Am. Chem. Soc.* **1982**, *104*, 7294–7299. See this reference for the details. In equation 1, the symbol eep (the ee of the product) was employed when ee0 = 0 and when ee0 > 0, instead of the symbol ee′, which was used in the reference when ee0 > 0.

(3) Chen, C. S.; Liu, Y. C. *J. Org. Chem.* **1991**, *56*, 1966–1968.

(4) Guo, Z. W.; Wu, S. H.; Chen, C. S.; Girdaukas, G.; Sih, C. J. *J. Am. Chem. Soc.* **1990**, *112*, 4942–4945.

(5) Mason, J. C. BASIC Numerical Mathematics; Butterworths: London, 1983; pp 66–67.

0022-3263/93/1958-5748$04.00/0

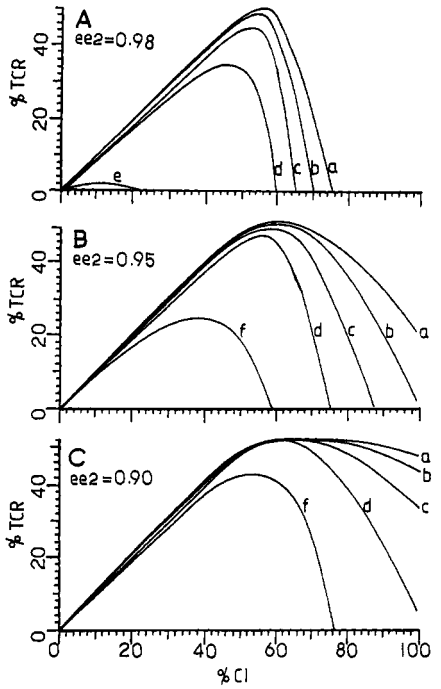

Figure 1. Expression of the percent theoretical chemical recovery (TCR = C1C2) as a function of the percent conversion of the first step (C1) for various fixed final product enantiomeric excesses (ee2) (A, 0.98; B, 0.95; C, 0.90) and enantiomeric ratios (E) (a, 50; b, 40; c, 30; d, 20; e, 11; f, 10) in two-step product recycling (ee0 = 0, $E_1 = E_2 = E$).

Two-Step Product Recycling. In this case, $n = 2$, ee0 = 0, $E_1 = E_2 = E$, and the chemical form of SP2 (PP1 was transformed into SP2 through a nonselective process) is the same as that of SP1. Curves generated with program C2SR are plotted in Figure 1, which provides an overview of the effect of E and C1 on the TCR for fixed values of ee2. Obviously, for a known E and a fixed ee2, the MCR and the corresponding C1 can either be determined from the corresponding theoretical curve or generated by the program for greater accuracy.

Resolution of (±)-1. (*S*)-(−)-1 (naproxol) possesses the same configuration as (*S*)-(+)-2-(6-methoxy-2-naphthyl)propionic acid (naproxen). Both compounds possess antiinflammatory, analgesic, and antipyretic activities, whereas the corresponding (*R*)-enantiomers do not. The antiinflammatory activity of naproxol is slightly greater than that of naproxen.[6] Considering that arylacetic acids commonly have side effects on the stomach and intestines, it is useful to study the enzymatic resolution of (±)-1. Hence, several commercially available lipases were tested, and enzymatic hydrolyses of various esters of (±)-1 and esterifications of (±)-1 with different acyl donors in organic media were attempted. Unfortunately, a highly enantioselective system was not found. In the best case, (±)-1 was esterified in vinyl acetate catalyzed by porcine pancreatic lipase (PPL) with an E value of 38 (±4),[7] as shown in Figure 2. It was a suitable example for two-step

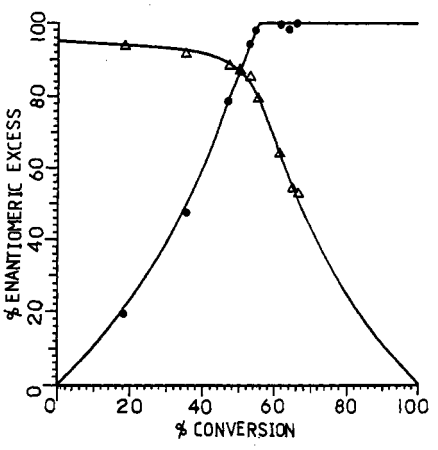

Figure 2. Enantioselective acylation of (±)-1 with vinyl acetate catalyzed by PPL. ● and ▲ were experimentally determined values of the enantiomeric excess of the substrate remaining (ees1) and the enantiomeric excess of the product (ee1), respectively. The curves were generated for $E = 38$ with program EEP and the relationship eesi = [Ci(eei − ee_{i-1})]/(1-Ci), which was derived from eq 3, where $i = 1$ and ee_{i-1} = ee0 = 0.

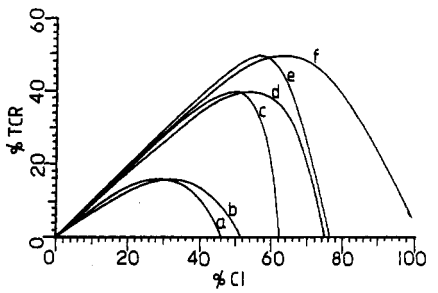

Figure 3. Expression of the percent theoretical chemical recovery (TCR = C1C2) as a function of the percent C1 for various fixed final product enantiomeric excesses (ee2) (a, b, 0.98; c, d, 0.95; e, f, 0.90) and E1, E2 (a, c, e, $E_1 = 20$, $E_2 = 10$; b, d, f, $E_1 = 10$, $E_2 = 20$) in general cases of combined two-step resolution for ee0 = 0.

recycling study, and the b curves ($E = 40$) in Figure 1 provide a general overview. For $E_1 = E_2 = 38$, ee0 = 0, and ee2 = 0.98, the MCR and the corresponding C1 and C2 were computer determined to be 49%, 54%, and 90%, respectively, with program C2SR. The program obviously did make useful predictions. Two-step recycling resolution experiments on the esterification of (±)-1 catalyzed by PPL in vinyl acetate were done in three trials. The three trials gave C1 values of 55%, 53%, and 53% and C2 values of 90%, 88%, and 92%; thus, the TCR values (equal to C1C2) for the three trials were 50%, 47%, and 49%, respectively; the isolated yield values for the whole process based on the amount of the starting racemic pair (equal to Y1 × Y2, see Experimental Section) were found to be 47%, 45%, and 48%, respectively, and the ee2 values were found to be 0.97, 0.99, and 0.98, respectively. The experimental data were in agreement with the predictions.

General Cases of Combined Two-Step Resolution. In a general case of combined two-step resolution, SP2 does not necessarily have the same chemical form as SP1, and, in addition, the second resolution step is not necessarily the same as the first. However, the quantitative analyses should be the same as those for two-step recycling case, except that $E_1 \neq E_2$. Figure 3 was generated with program C2SR for a combined two-step resolution with E_1 and E_2 set at 10 and 20, respectively. It is interesting that the MCR in the case in which $E_1 = 10$ and $E_2 = 20$

(6) Harrison, I. T.; Lewis, B.; Nelson, P.; Rooks, W.; Roszkowski, A.; Tomolonis, A.; Fried, J. H. *J. Med. Chem.* **1970**, *13*, 203–205.

(7) The calculation of E was based on data for six samples with C1 values of 18%, 35%, 47%, 50%, 53%, and 55%, and the E values obtained were 34, 34, 38, 41, 43, and 38, respectively. Equation 6, which was derived from eqs 2 and 3, where $i = 1$, was used for the calculation of E.

$$E_i = \frac{\ln [(1 - \text{eesi})(\text{eei} - \text{ee}_{i-1})/[(\text{eesi} + \text{eei})(1 + \text{ee}_{i-1})]]}{\ln [(1 + \text{eesi})(\text{eei} - \text{ee}_{i-1})/[(\text{eesi} + \text{eei})(1 - \text{ee}_{i-1})]]} \quad (6)$$

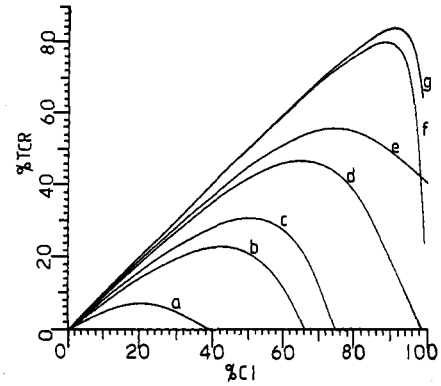

Figure 4. Expression of the percent theoretical chemical recovery (TCR = C1C2) as a function of the percent C1 for various known enantiomeric excesses of the starting substrate (ee0) and fixed final product enantiomeric excesses (ee2) for the preparation of the (*S*)-isomer by coupled enantioselective action on antipodal 2, where $E_1 = 5.6$ and $E_2 = 9.8$. The data given below in brackets for each curve are ee0, ee2, MCR (maximal chemical recovery), and the corresponding C1, respectively; a (0, 0.95, 6%, 21%); b (0, 0.92, 23%, 43%); c (0, 0.90, 30%, 50%); d (0, 0.82, 47%, 65%); e (0, 0.72, 56%, 74%); f (0.82, 0.98, 79%, 88%); g (0.72, 0.95, 84%, 91%).

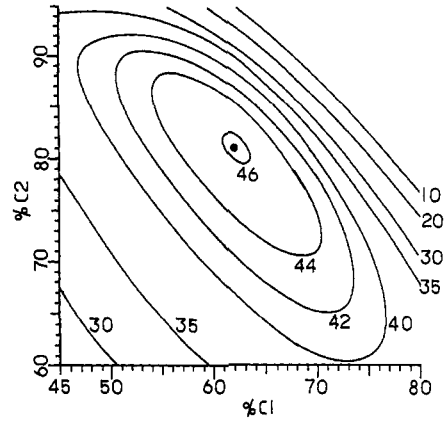

Figure 5. Plot of the percent theoretical chemical recovery (TCR = C1C2C3) over two dimensions (%C1 and %C2) for combined three-step resolution when ee0 = 0, $E_1 = E_2 = E_3 = 10$, and the final product enantiomeric excess (ee3) was fixed at 0.95. The number next to each curve denotes the percent TCR. The point at the center denotes the maximal chemical recovery (MCR = 46%) and the corresponding conversions of the first two steps (C1 = 62%, C2 = 81%). The corresponding conversion of the final step (C3) should be equal to MCR/(C1C2) = 92%.

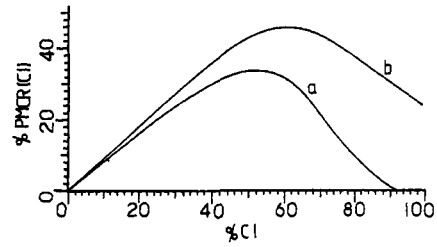

Figure 6. Expression of the percent partial maximal chemical recovery (the corresponding C1C2C3) for a fixed C1 (PMCR(C1)) as a function of the percent C1 for various fixed final product enantiomeric excesses (ee3) (a, 0.98; b, 0.95) in combined three-step resolution when ee0 = 0, $E_1 = E_2 = E_3 = 10$. Data given below in brackets for each curve are maximal chemical recovery (MCR) of the whole three-step process and the corresponding C1, C2, and C3, respectively: a (34%, 53%, 76%, 85%); b (46%, 62%, 81%, 92%).

was the same as that in the case in which $E_1 = 20$ and $E_2 = 10$ when ee2 was fixed at the same value for each case. However, the first-step reactions had to be terminated at different conversion values for each case to reach the MCR of the whole two-step resolution process.

Coupled Enantioselective Action. Figure 4, generated with program C2SR, was based on an experimental example named coupled enantioselective action by Chen.[3] In the experiments, the racemate (ee0 = 0) of 2-phenyl-*n*-propyl butyrate (**2**) was deacylated with butanol in hexane catalyzed by PPL with a selectivity of $E_1 = 5.6$ (determined previously) with a preference for the (*S*)-enantiomer. An excess of an achiral acyl donor (vinyl acetate) was added to the above system midway through the reaction; the alcohol intermediate that had been partly optically enriched (ee1) underwent acylation with vinyl acetate catalyzed by the same enzyme with a selectivity of $E_2 = 9.8$ (determined previously) with the same preference for the (*S*)-enantiomer. Under the assumption of the absence of deacylation in the presence of an excess of the achiral acyl donor, the quantitative analysis for the coupled resolution action should be the same as that for a general case of combined two-step resolution. Figure 4 shows that the first step should be stopped by the addition of vinyl acetate when C1 = 21%, 43%, 50%, 65%, and 74% in order to reach the MCR when ee2 was fixed at 0.95, 0.92, 0.90, 0.82, and 0.72, respectively.

Computer Program C3SR. Program C3SR was compiled for quantitative analyses of combined three-step resolutions. E_1, E_2, E_3, and ee0 were determined, and ee3 was fixed at an appropriate value. Then C3 was made a numerical function of C1 and C2 by sequentially using program EEP for step 1 and step 2 and program C for step 3. TCR, equal to C1C2C3, was also made a numerical function of C1 and C2. MCR and the corresponding C1, C2, and C3 were determined by the program. The plot of TCR over two dimensions (C1 and C2) could be drawn (Figure 5). By fixing C1, TCR was made a numerical function of C2 only, and the partial MCR for fixed C1 (symbolized on PMCR(C1)) could be determined. Theoretical curves of PMCR(C1) against C1 could be plotted (Figure 6). The MCR and the corresponding C1 could also be determined in this way.

Cases when $n \geq 4$. In principle, for a combined *n*-step resolution, the TCR, equal to C1C2···C*n*−1C*n*, could be made a numerical function of C1, C2, ···, C*n*−1 by sequentially using program EEP for steps 1, 2, ···, and *n*−1 and program C for step *n*. The MCR and the corresponding C1, C2, ···, C*n* could be determined by computer. However, no easy-view plot of the TCR over three or more dimensions could be drawn. Herein, an alternative method using program C2SR and C3SR repeatedly is disclosed. For example, if it is necessary to obtain (*S*)-**2** with a very high ee, a possible method is to do the coupled action twice. This method is a case of combined four-step resolution, where ee0 = 0, $E_1 = E_3 = 5.6$, $E_2 = E_4 = 9.8$. The correlation of the two coupled actions is made with ee2. For a given ee2, a maximal C1C2 and the corresponding C1 and C2 could be determined for the first coupled action, and, similarly, a maximal C3C4 and the corresponding C3 and C4 could be determined for the second when the final product enantiomeric excess (ee4) was fixed. Thus, the partial maximal C1C2C3C4 (symbolized as PMCR(ee2)) was made a numerical function of ee2. Then, the MCR for the whole process and the corresponding C1–C4 could

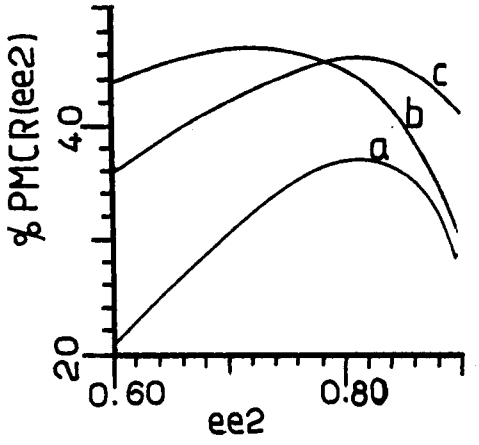

Figure 7. Expression of the percent partial maximal chemical recovery (the corresponding C1C2C3C4) for a given ee2 (PMCR(ee2)), as a function of ee2 for various fixed final product enantiomeric excesses (ee4) and E_i (i = 1–4) in combined four-step resolution when ee0 = 0. Data given below in brackets for each curve are E_1, E_2, E_3, E_4, ee4, maximal chemical recovery (MCR) of the whole four-step process, and the corresponding ee2, C1, C2, C3, and C4, respectively: a (5.6, 9.8, 5.6, 9.8, 0.98, 37%, 0.82, 65%, 72%, 88%, 90%); b (5.6, 9.8, 5.6, 9.8, 0.95, 47%, 0.72, 74%, 75%, 91%, 92%); c (10, 10, 10, 10, 0.98, 46%, 0.82, 64%, 82%, 92%, 95%).

be determined. Curves a and b in Figure 7 were generated when ee4 was fixed at 0.98 and 0.95, respectively. Curve c in Figure 7 corresponds to a four-step product recycling case, in which ee0 = 0, $E_1 = E_2 = E_3 = E_4 = 10$, and ee4 was fixed at 0.98.

Conclusions

If a desired enantiomer is the substrate remaining after an irreversible enantioselective reaction, it is easy to obtain useful amounts of the desired enantiomer with high enantiomeric purity through one enzymatic reaction step, even a moderately selective one (e.g., E = 10), by carrying the reaction to higher conversion (e.g., 70% when E = 10).[2] However, if the desired enantiomer is the preferred isomer of an enzyme with only moderate E, it is impossible to obtain the desired isomer with high enantiomeric purity by a single resolution step. In this work, combined multistep resolutions including the typical product recycling,[2] the coupled enantioselective action,[3] and other combinations of multiple resolution steps (e.g., enzymatic esterification followed by enzymatic hydrolysis catalyzed either by the same lipase or different lipases with the same enantiomeric preference) were considered. For any designed combined n-step resolution, useful predictions of the MCR of the desired enantiomer with a fixed final product enantiomeric excess (een) and the corresponding conversions of all steps were determined with both new calculation methods and novel plots. This work should encourage chemists to prepare desired enantiomers with high optical purity by using even moderately enantioselective enzymatic reactions. A large number of enzymes exist for many unnatural substrates, and they are easy to handle. Commercially available lipases are especially useful for preparing optically active alcohols and acids. When an enzyme with E = 10 is used in product recycling with ee0 = 0, the MCR is 24% for a combined two-step resolution when ee2 is fixed at 0.95 (curve f in Figure 1B), 46% for a combined three-step resolution when ee3 is fixed at 0.95 (Figure 5 or curve b in Figure 6), 34% for a combined three-step resolution when ee3 is fixed at 0.98 (curve a in Figure 6), and 46% for a combined four-step resolution when ee4 is fixed at 0.98 (curve c in Figure 7).

For nonenzymatic kinetic resolution under pseudo-first-order conditions, the relative rate difference (k_{fast}/k_{slow}) adopted by Sharpless[8] or the stereoselectivity factor (S) used by Kagan[9] can be employed instead of E for enzymatic resolution. S and k_{fast}/k_{slow} are similar to the biochemical stereoselectivity factor (E) for quantitative analyses, although E depends on the ratio of the enzymatic specificity constants. Thus, all the plotting methods disclosed in this work are also applicable to nonenzymatic combined multistep resolutions for making useful quantitative predictions.

Experimental Section

The chiral shift reagent Eu(hfc)$_3$ was obtained from Aldrich Chemical Co. PPL was a type II product of Sigma Chemical Co. and had a specific activity of 54 olive oil units per mg of solid. (±)-1 was prepared from (±)-2-(6-methoxy-2-naphthyl)propionic acid. All other chemicals and solvents used in this work were of analytical grade, and all solvents were glass-distilled prior to use. Quantitative analyses were performed on an IBM-AT-286 personal computer using BASIC programs written by the author.

Enantioselective Acylation of (±)-1. PPL powder was added to a clear solution of (±)-1 (SP1) in vinyl acetate. The ratio of (±)-1/vinyl acetate/PPL was 1 mmol/4 mL/200 mg. The mixture was incubated on a rotary shaker (200 rpm) at 25 °C for the indicated reaction period. The reaction was terminated by filtering off the lipase powder and washing it with ethyl acetate. The organic solution was evaporated to dryness under reduced pressure, and the residue was chromatographed on a silica gel (120–160 mesh) column with hexane–ethyl acetate (5/1–1/1) to afford the product acetate of (*S*)-1 (PP1) and the remaining (*R*)-1 (RP1) for ees1 determination. PP1 was subjected to alkaline hydrolysis (2 N NaOH/MeOH, 60 °C, 3 h) to yield (*S*)-1 (SP2) for ee1 determination. The isolated yield (*Y*1) of (*S*)-1 for this step was determined based on the amount of the starting racemic pair. The procedure for ee determination is described below. The conversion could be determined with eq 3, where i = 1 and ee_{i-1} = ee0 = 0. For 1, 2, 4, 8, 14, 16, 24, 48, and 120 h, the C1 values were found to be 18%, 35%, 47%, 50%, 53%, 55%, 61%, 64%, and 66%, respectively; the ee1 values were 0.93, 0.91, 0.88, 0.87, 0.85, 0.79, 0.64, 0.54, and 0.52, respectively; the ees1 values were 0.20, 0.48, 0.79, 0.87, 0.94, 0.98, 1.0, 0.98, and 1.0, respectively. The E value was calculated[7] to be 38 (±4).

Product Recycling Step. PPL powder (200 mg) was added to a clear solution of SP2 (216 mg, 1.0 mmol, antipodal alcohol 1, obtained from the product of the first resolution step with ee1) in vinyl acetate (4 mL). The reaction conditions and the workup procedure were similar to those described above and yielded the acetate of (*S*)-1 (PP2) and recovered substrate (*R*)-1 (RP2) for ees2 determination. PP2 was subjected to alkaline hydrolysis (2 N NaOH/MeOH, 60 °C, 3 h) to yield (*S*)-1 (SP3) for ee2 determination. The isolated yield (*Y*2) of (*S*)-1 for this second step was determined based on the amount of SP2, and then the isolated yield of (*S*)-1 for the whole two-step product recycling process including two enzymatic acylation steps and two nonselective alkaline hydrolysis steps was calculated to be $Y1 \times Y2$. C2 was determined with eq 3, where i = 2 and ee_{i-1} = ee1 > 0. For the three trials, the ee1 values were 0.79, 0.85, and 0.85; the reaction times were 12, 12, and 13 h, respectively; the ee2 values

(8) Martin, V. S.; Woodard, S. S.; Katsuki, T.; Yamada, Y.; Ikeda, M.; Sharpless, K. B. *J. Am. Chem. Soc.* **1981**, *103*, 6237–6240.
(9) Kagan, H. B.; Fiaud, J. C. *Top. Stereochem.* **1988**, *18*, 249–330.

were 0.97, 0.99, and 0.98, respectively; the ees2 values were 0.87, 0.16, and 0.74, respectively; C2 values were 90%, 88%, and 92%, respectively; and Y2 values were 88.8%, 86.0%, and 91.9%, respectively. Thus, C1 × C2 values were 50%, 47%, and 49%, respectively, and Y1 × Y2 values were 47%, 45%, and 48%, respectively.

Determination of ee. The ee was determined by means of the rotation of antipodal alcohol 1 in chloroform (ee = $|[\alpha]_D/18|$)[6] and was confirmed by 200-MHz ^{1}H NMR analysis in the presence of the chiral shift reagent Eu(hfc)$_3$. For NMR analysis, each sample contained 8 mg of antipodal alcohol 1, 16 mg of Eu(hfc)$_3$, and 0.5 mL of $CDCl_3$. The apparent singlet (δ 7.60, J = 1.8 Hz) for 1-H on the naphthyl ring was shifted to about δ 10 and split into two singlets. For example, the signals of (*S*)-(−)-1 and (*R*)-(+)-1 appeared at δ 10.11 and 10.03, respectively. The ee of each sample was calculated according to the ratio of the integral areas of the signals for each of the two enantiomers, and these ee were in agreement with the optical rotation data.

Acknowledgment. This work was supported by the National Foundation of Natural Sciences.

Abbreviations: *E*, enantiomeric ratio; eep, ee of the product of an enzymatic reaction (see ref 2); SP, starting substrate pair; PP, product pair; RP, substrate remaining pair; *i*, serial number of each resolution step; eei, ee of fast-reacting enantiomer of PPi (the same as of SP_{i+1}); eesi, ee of slow-reacting enantiomer of RPi; Ci, conversion of step *i*; TCR, theoretical chemical recovery; MCR, maximal chemical recovery; PMCR(C1), partial maximal chemical recovery for a fixed C1; PMCR(ee2), partial maximal chemical recovery for a given ee2; *Y*, isolated yield.