Available online at www.sciencedirect.com
SCIENCE DIRECT
Tetrahedron
Tetrahedron 62 (2006) 2912–2916

Enantio-complementary deracemization of (±)-2-hydroxy-4-phenylbutanoic acid and (±)-3-phenyllactic acid using lipase-catalyzed kinetic resolution combined with biocatalytic racemization

Barbara Larissegger-Schnell,[a] Silvia M. Glueck,[b] Wolfgang Kroutil[a] and Kurt Faber[a,*]

[a]*Department of Chemistry, Organic and Bioorganic Chemistry, University of Graz, Heinrichstrasse 28, A-8010 Graz, Austria*
[b]*Research Centre Applied Biocatalysis, Graz, Austria*

Received 25 November 2005; revised 20 December 2005; accepted 4 January 2006

Available online 20 January 2006

Abstract—Deracemization of (±)-3-phenyllactic acid (**1**) and (±)-2-hydroxy-4-phenylbutanoic acid (**2**) was accomplished by lipase-catalysed kinetic resolution coupled to biocatalytic racemization of the non-reacting substrate enantiomers using *Lactobacillus paracasei* DSM 20008. Cyclic repetition of this sequence led to a single enantiomeric product from the racemate. Access to both enantiomers was achieved by switching between lipase-catalysed acyl-transfer and ester hydrolysis reactions. Both products constitute important building blocks for virus protease- and ACE-inhibitors, respectively.

1. Introduction

3-Phenyllactate (**1**) and derivatives thereof are frequently used in nonracemic form as components of pharmaceuticals and natural antibiotic agents.[1] They represent an integral part of bioactive peptides, such as Aeruginosins[2] and Microcin,[3] which were shown to be potent protease inhibitors. The *p*-fluoro-analog of **1** is a key building block for the synthesis of AG7088 (Ruprintrivir), a potent rhinovirus inhibitor currently being tested in clinical trials to treat the common cold.[4] Several approaches to obtain **1** in nonracemic form have been reported.[5] The majority of them is based on racemate resolution[6] or the asymmetric transformation of a suitable non-chiral synthetic precursor.[4a,7]

(*R*)-2-Hydroxy-4-phenylbutanoic acid (**2**) is an important building block for the production of a large variety of angiotensin converting enzyme (ACE) inhibitors having in common the (*S*)-homophenylalanine moiety as the central pharmacophore unit.[8] These agents from the the 'pril-family', such as Enalapril, Lisinopril, Cilapril or Benazepril, efficiently expand the range of antihypertensives, like β-blockers, A_2-antagonists or Ca-channel blockers. Due to the fact that many of these drugs have lost patent protection (or soon will do so), the production costs of the required building blocks has become a major issue. For the synthesis of **2** in nonracemic form, numerous strategies based on (i) racemate resolution via crystallization[9] or (ii) kinetic resolution of a racemate[10] or (iii) the asymmetric transformation of a prochiral precursor[11] have been devised.

The majority of these routes have one or more weak points:[4,5,11m] high cost of reagents, such as chiral transition metal complexes, insufficient catalyst selectivity or activity, sensitivity of catalysts in asymmetric hydrogenation, or low stability of starting materials, such as α-keto acids.

The most dramatic limitation common for all strategies relying on kinetic resolution is the maximum theoretical yield of 50% for a single enantiomer. In order to overcome this fundamental drawback, two approaches—summarized under the term 'deracemization'[12]—were recently proposed: (i) dynamic kinetic resolution[13] and (ii) microbial stereo-inversion.[14]

Although both of these methods have the clear merit of a 100% theoretical yield of a single stereoisomeric product, its absolute configuration is determined by the enantiopreference of the biocatalyst employed. Since mirror-image enzymes *sensu stricto* do not exist,[15] production of both

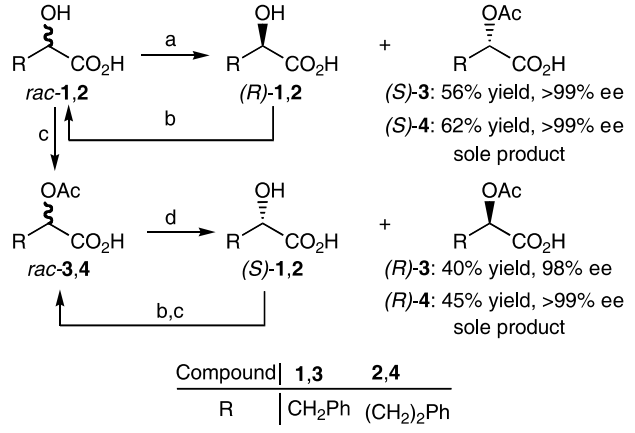
Keywords: Enantio-complementary; Deracemization; Lipase; Biocatalytic racemization; Biocatalysis.
* Corresponding author. Tel.: +43 316 380 5332; fax: +43 316 380 9840; e-mail: kurt.faber@uni-graz.at

0040–4020/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tet.2006.01.007

enantiomers through biocatalytic deracemization by simple choice of the 'matching enantiomer' of the chiral (bio)catalyst is virtually impossible.

In order to circumvent this limitation, we envisaged to apply lipase-catalyzed ester hydrolysis and ester formation via acyl transfer to our recently developed deracemization protocol[16] based on the enzymatic racemization of the non-reacting substrate enantiomer using a racemase.[17] Taking into consideration that ester hydrolysis and esterification represent reactions in opposite directions, products of opposite configuration are to be expected. The proof of principle for this concept was recently verified by us using a lipase–mandelate racemase two-enzyme process.[18] However, the restricted substrate tolerance of mandelate racemase (EC 5.1.2.2), which is unable to isomerize 3-phenyllactate (**1**) or 2-hydroxy-4-phenylbutanoate (**2**) imposed severe limitations[19] and forced us to employ a suitable β,γ-unsaturated synthetic precursor for 2-hydroxy-4-phenylbutanoate (i.e., 2-hydroxy-4-phenylbut-3-enoate) instead, which required additional synthetic steps and thus unfavorably enhanced the overall complexity of the process. Since this substrate-analog-trick could not be applied to 3-phenyllactate (**1**), the latter compound could not be deracemized by using the above mentioned mandelate racemase–lipase protocol. In search for a suitable biocatalyst for the racemization of a wide variety of saturated aliphatic 2-hydroxycarboxylic acids, we recently identified whole resting cells of *Lactobacillus* sp. as a promising alternative.[20]

In order to demonstrate the general applicability of this method, we envisaged to develop an enantio-complementary deracemization protocol as follows (Scheme 1).



Scheme 1. Reagents and conditions: (a) *Pseudomonas* sp. lipase (Amano PS-C-II), vinyl acetate, *i*-Pr_2O, 25 °C; (b) *Lactobacillus paracasei* DSM 20008, buffer pH 6.5, 42 °C; (c) Ac_2O, pyridine; (d) porcine pancreas lipase (EC 3.1.1.3, for **3**), *Candida antarctica* lipase B/Novozyme 435 (for **4**), buffer pH 7.5, acetone, 30 °C.

2. Results and discussion

2.1. (*S*)-Series (Scheme 1)

Kinetic resolution of (±)-**1** via *Pseudomonas* sp. lipase catalysed acyl-transfer using vinyl acetate as acyl donor proceeded with excellent enantioselectivities ($E>200$) to furnish a mixture of (*S*)-**3** and unreacted (*R*)-**1** in >99% ee at 50% conversion. Biocatalytic racemization of (*R*)-**1** was accomplished without separation from the *O*-acetylated product **3** using whole cells of *Lactobacillus paracasei* DSM 20008 in aqueous buffer. After the mixture of *rac*-**1** and (*S*)-**3** thus obtained was subjected to two subsequent cycles of lipase-catalysed kinetic resolution/biocatalytic racemization as described above, (*S*)-**3** was obtained in 56% overall yield from the racemate as the sole product. Finally, hydrolysis of the *O*-acetyl group gave (*S*)-3-phenyllactic acid (**1**) without loss of optical purity in 69% yield and >99% ee.

When this deracemization protocol was applied to 2-hydroxy-4-phenylbutanoic acid (*rac*-**2**), (*S*)-**2** was obtained in >99% ee in similar yield.

2.2. (*R*)-Series (Scheme 1)

It was expected, that lipase-catalysed hydrolysis of *rac*-*O*-acetate esters **3** and **4** would give access to the corresponding enantiomeric products, that is, (*S*)-**1**,**2** and nonreacted α-acetoxycarboxylic acids (*R*)-**3**,**4**. However, initial attempts were hampered by solubility problems of the rather polar substrates and insufficient enantioselectivities of various lipases. For instance, *Candida rugosa* lipase (Amano AY), various *Pseudomonas* sp. lipases (Amano PS-C-I, PS-D), and *Candida antarctica* lipase A displayed low enantioselectivity. After some experimentation, we identified two lipases, that is, porcine pancreas lipase (EC 3.1.1.3) for *rac*-**3** and *Candida antarctica* lipase B for *rac*-**4**, which showed excellent enantioselectivity. In addition, solubility problems were overcome by using acetone as co-solvent. Without separation of materials, the cyclic sequence of acetylation/hydrolysis/racemization was repeated two times to give (*R*)-**3** and (*R*)-**4** in 98% and >99% ee, respectively, as the sole products. Finally, deacylation of the latter compounds furnished α-hydroxycarboxylic acids (*R*)-**1** and (*R*)-**2** in 74–75% yield. Biocatalysts could be recovered by filtration and reused for the whole number of cycles until completion of the process.

The main limitation of this process lies in the neccessity to switch between aqueous-organic solvent systems, which furnishes a consecutive sequence of steps rather than a dynamic process. Attempts to conduct the biocatalytic racemization in situ in an organic solvent were unsuccessful so far. Although isolated overall yields were still below the theoretical 100%, this threshold should be approachable using improved recovery procedures for the polar hydroxycarboxylic acids from the aqueous medium (e.g., by ion exchange chromatography or continuous extraction). Detailed analysis showed that this process is virtually free of yield-limiting side reactions and that microbial degradation of *rac*-**1**,**2** during the racemization step by was negligible.[21] The possibility to determine the stereochemical configuration of the sole product by a simple switch between lipase-catalyzed acyl-transfer- and hydrolysis-mode demonstrates the flexibility of this process.

3. Conclusion

In summary, both enantiomers of 3-phenyllactic acid (**1**) and 2-hydroxy-4-phenylbutanoic acid (**2**) were obtained as single stereoisomers in >98% ee from the corresponding racemates via a stepwise deracemization protocol consisting of a lipase-kinetic resolution followed by biocatalytic racemization. Whereas (*S*)-enantiomers were accessed through lipase-catalysed acyl-transfer, the (*R*)-counterparts were obtained via ester hydrolysis.

4. Experimental

4.1. General

Determination of conversion and enantiomeric excess were determined via HPLC on a chiral stationary phase. HPLC analyses were carried out on a Jasco HPLC-system (pumps PU-980, multi-wave-length-detector MD-910, autosampler AS-950, degasser CMA/260) using a Chiralpak AD column (column A, Daicel, 0.46 cm×25 cm). Compounds were purified by flash chromatography on silica gel Merck 60 (230–400 mesh). Melting points were obtained on a Gallenkamp melting point apparatus MFB-595 in open capillary tubes, optical rotation values were measured on a Perkin-Elmer polarimeter 341 at 589 nm (Na-line) in a 1 dm cuvette and are given in units of 10^{-1} deg cm^2 g^{-1}.

4.1.1. (*S*)-*O*-Acetyl-3-phenyllactic acid (*S*)-3. Kinetic resolution step (a): to a solution of *rac*-**1** (0.5 g, 3 mmol) in diisopropyl ether (50 mL), vinyl acetate (5 mL) and *Pseudomonas* sp. lipase (Amano PS-C-II, 0.5 g) were added and the mixture was shaken for 48 h at 25 °C and 150 rpm. The enzyme was filtered, washed and dried for reuse, the filtrate was evaporated to dryness.

Racemization step (b): (*S*)-**3** and (*R*)-**1** obtained from step (a) were dissolved in bis-Tris-buffer (5 mL, 50 mmol, pH 6.5, 10^{-2} M $MgCl_2$) and the pH was adjusted to 6.5. Then, lyophilized whole cells of *Lactobacillus paracasei* DSM 20008[20b] [2 g, rehydrated in bis-Tris-buffer (12 mL, 50 mmol, pH 6.5, 10^{-2} M $MgCl_2$) for 30 min] were added and the mixture was shaken for 48–72 h at 42 °C and 120 rpm until racemization of (*R*)-**1** was complete. After centrifugation, the solution was acidified to pH 1–2 with HCl (3 M) and *rac*-**1** and (*S*)-**3** were extracted three times with ethyl acetate, dried with Na_2SO_4 and evaporated.

After cyclic repetition of step (a) twice and step (b) once, the residue obtained by extraction was purified by flash chromatography (to remove minor impurities emerging from the cells) using dichloromethane/methanol (gradient from 0–10% MeOH) to yield (*S*)-**3** as the sole product (0.35 g, 56%, oil); HPLC analysis showed a single peak at T_{Ret} 14.8 min using a Chiralpak AD column (Daicel, heptane/2-propanol/CF_3COOH 90:10:0.1; 0.5 mL/min, 18 °C); $[\alpha]_D^{20}$ −10.25 (*c* 1.79; acetone, >99% ee).

4.1.2. (*S*)-3-Phenyllactic acid (*S*)-1. Hydrolysis step: a mixture of (*S*)-**3** (0.2 g, 1 mmol), MeOH (8 mL) and K_2CO_3 (1 g) was stirred at 0 °C for 5 h. After acidification with HCl (3 M) to pH 1–2, the product was extracted three times with ethyl acetate, the organic layer was dried (Na_2SO_4), evaporated and the residue was purified by flash chromatography using dichloromethane/methanol (gradient from 0–10% MeOH) as eluent to yield (*S*)-**1** (0.11 g, 69%); mp 122–123 °C; lit.[22] mp: 120–121 °C; $[\alpha]_D^{20}$ −27.55 (*c* 1.0; acetone, >99% ee); lit.[23] $[\alpha]_D^{25}$ −27.80 (*c* 1.13; acetone); HPLC analysis using the method described above showed a single peak at T_{Ret} 24.53 min.

4.1.3. (*S*)-2-Acetoxy-4-phenylbutanoic acid (*S*)-4. In the same manner as described above, *rac*-**2** (0.5 g, 2.77 mmol) was converted to (*S*)-**4** (0.38 g) in a total yield of 62%; mp: 28–30 °C; $[\alpha]_D^{20}$ −6.61 (*c* 0.5; acetone, >99% ee); lit.[24] $[\alpha]_D^{25}$ −11.0 (*c* 0.92; EtOH); HPLC analysis showed a single peak at T_{Ret} 19.12 min using a Chiralpak AD column (Daicel, heptane/2-propanol/CF_3COOH 90:10:0.1; 0.4 mL/min, 18 °C).

4.1.4. (*S*)-2-Hydroxy-4-phenylbutanoic acid (*S*)-2. In the same manner as described above, (*S*)-**4** (0.22 g, 1 mmol) was converted to (*S*)-**2** (0.13 g; 73%); mp 115–117 °C; lit.[23] mp: 114 °C; $[\alpha]_D^{20}$ +8.1 (*c* 1.0; EtOH, >99% ee); lit.[23] $[\alpha]_D^{25}$ +7.5 (*c* 0.5; EtOH, 84% ee). HPLC analysis using the method described above showed a single peak at T_{Ret} 27.01 min.

4.1.5. (*R*)-*O*-Acetyl-3-phenyllactic acid (*R*)-3. Acylation step (c): a solution of *rac*-**1** (0.5 g, 3 mmol) in acetic anhydride (4 mL) and pyridine (0.25 mL) was kept at 0–5 °C. After 6 h the solution was poured into ice-water (50 mL), which was acidified with HCl (3 M) to pH 1–2 and extracted three times with ethyl acetate. The combined organic layers were washed with H_2O and brine, dried (Na_2SO_4) and evaporated to yield *rac*-**3** (0.50 g, 80%).

Kinetic resolution step (d): to a solution of *rac*-**3** (0.5 g, 2.6 mmol) in acetone (21 mL) and phosphate buffer (35 mL, pH 7.5; 50 mmol), lipase from porcine pancreas (EC 3.1.1.3, 5 g, Sigma Type II, crude) was added and the mixture was shaken for 20 h at 30 °C and 150 rpm. After centrifugation, acetone was evaporated from the filtrate. The residue was acidified with HCl (3 M) to pH 1–2, extracted three times with ethyl acetate, dried (Na_2SO_4) and evaporated.

For the racemization step (b) see above.

After performing steps (c) and (d) twice and step (b) once, the residue obtained by extraction was purified by flash chromatography (to remove minor impurities emerging from the cells) using dichloromethane/methanol (gradient from 0–10% MeOH) to yield (*R*)-**3** as the sole product (0.25 g, 40%), oil; $[\alpha]_D^{20}$ +8.86 (*c* 1.39; acetone, 98% ee). HPLC analysis using the method described above showed a single peak at T_{Ret} 17.80 min.

4.1.6. (*R*)-3-Phenyllactic acid (*R*)-1. In the same manner as described for (*S*)-**1**, (*R*)-**3** (0.15 g, 0.7 mmol) was converted to (*R*)-**1** (0.09 g, 75%); mp 124–125 °C; lit.[22] mp: 120–121 °C; $[\alpha]_D^{20}$ +26.95 (*c* 1.0; acetone, 98% ee); lit.[22] $[\alpha]_D^{25}$ 29.8 (*c* 1.13; acetone). HPLC analysis using the method described above showed a single peak at T_{Ret} 20.38 min.

4.1.7. (*R*)-2-Acetoxy-4-phenylbutanoic acid (*R*)-4. In the same manner as described above, *rac*-**2** (0.5 g, 2.8 mmol) was converted to *rac*-**4** (0.84 g, 87%).

Kinetic resolution step (d): to a solution of *rac*-**4** (0.54 g, 2.4 mmol) in acetone (2.5 mL) and phosphate buffer (25 mL, pH 7.5, 50 mmol), lipase from *Candida antarctica* B (Novozyme 435, 0.5 g) was added and the mixture was shaken for 24 h at 30 °C and 150 rpm. The lipase was recovered by filtration and dried for reuse. The filtrate was evaporated from acetone, the residue was acidified with HCl (3 M) to pH 1–2, extracted three times with ethyl acetate, dried (Na_2SO_4) and evaporated.

For the racemization step (b) see above.

After repeating steps (c) and (d) twice and step (b) once, the residue obtained by extraction was purified by flash chromatography (to remove minor impurities emerging from the cells) using dichloromethane/methanol (gradient from 0–10% MeOH) to yield (*R*)-**4** as sole product (0.28 g, 45%); mp: 27–30 °C; $[\alpha]_D^{20}$ +5.21 (*c* 1.36; acetone, >99% ee). HPLC analysis using the method described above showed a single peak at T_{Ret} 24.4 min.

4.1.8. (*R*)-2-Hydroxy-4-phenylbutanoic acid (*R*)-2. In the same manner as described above, (*R*)-**4** (0.2 g; 0.9 mmol) was converted to (*R*)-**2** (0.12 g, 74%); mp 115–117 °C; lit.[23] mp: 114 °C; $[\alpha]_D^{20}$ −8.5 (*c* 1.0; EtOH, >99% ee); lit.[23] $[\alpha]_D^{25}$ −9.0 (*c* 0.1; EtOH). HPLC analysis using the method described above showed a single peak at T_{Ret} 24.8 min.

Acknowledgements

This study was performed within the Research Centre Applied Biocatalysis. Financial support by BASF AG (Ludwigshafen), TIG, FFG, Province of Styria and City of Graz is gratefully acknowledged. B. Hauer and R. Stürmer (BASF-AG) are thanked for their valuable contributions.

References and notes

1. (a) Ström, K.; Sjögren, J.; Broberg, A.; Schnürer, J. *Appl. Environ. Microbiol.* **2002**, *68*, 4322–4327. (b) Dieuleveux, V.; van der Pyl, D.; Chataud, J.; Gueguen, M. *Appl. Environ. Microbiol.* **1998**, *64*, 800–803.
2. (a) Valls, N.; Lopez-Canet, M.; Vallribera, M.; Bonjoch, J. *Chem. Eur. J.* **2001**, *7*, 3446–3460. (b) Valls, N.; Vallribera, M.; Carmeli, S.; Bonjoch, J. *Org. Lett.* **2003**, *5*, 447–450. (c) Ishida, K.; Okita, Y.; Matsuda, H.; Okino, T.; Murakami, M. *Tetrahedron* **1999**, *55*, 10971–10988.
3. Valls, N.; Vallribera, M.; Lopez-Canet, M.; Bonjoch, J. *J. Org. Chem.* **2002**, *67*, 4945–4950.
4. (a) Tao, J.; McGee, K. *Org. Process Res. Dev.* **2002**, *6*, 520–527. (b) Dragovich, P. S.; Prins, T. J.; Zhou, R.; Brown, E. L.; Maldonado, F. C.; Fuhrman, S. A.; Zalman, L. S.; Tuntland, T.; Lee, C. A.; Patick, A. K.; Matthews, D. A.; Hendrickson, T. F.; Kosa, M. B.; Liu, B.; Batugo, M. R.; Gleeson, J.-P. R.; Sakata, S. K.; Chen, L.; Guzman, M. C.; Meador, J. W.; Ferre, R. A.; Worland, S. T. *J. Med. Chem.* **2002**, *45*, 1607–1623.
5. Storz, T.; Dittmar, P.; Fauquex, P. F.; Marschal, P.; Lottenbach, W. U.; Steiner, H. *Org. Process Res. Dev.* **2003**, *7*, 559–570.
6. Adam, W.; Lazarus, M.; Schmerder, A.; Humpf, H.-U.; Saha-Möller, C. R.; Schreier, P. *Eur. J. Org. Chem.* **1998**, 2013–2018.
7. Aladro, F. J.; Guerra, F. M.; Moreno-Dorado, F. J.; Bustamante, J. M.; Jorge, Z. D.; Massanet, G. M. *Tetrahedron Lett.* **2000**, *41*, 3209–3213.
8. (a) Sheldon, R. A. *Chirotechnology*; Marcel Dekker: New York, 1993; pp 362–367. (b) Sheldon, R. A.; Zeegers, H. J. M.; Houbiers, J. P. M.; Hulshof, L. A. *Chimica Oggi* **1991**, *9*, 35–47.
9. Attwood, M. R.; Hassall, C. H.; Kröhn, A.; Lawton, G.; Redshaw, S. *J. Chem. Soc., Perkin Trans. 1* **1986**, 1011–1019.
10. (a) Liese, A.; Kragl, U.; Kierkels, H.; Schulze, B. *Enzyme Microb. Technol.* **2002**, *30*, 673–681. (b) Kalaritis, P.; Regenye, R. W.; Partridge, J. J.; Coffen, D. L. *J. Org. Chem.* **1990**, *55*, 812–815. (c) Chadha, A.; Manohar, M. *Tetrahedron: Asymmetry* **1995**, *6*, 651–652. (d) Osprian, I.; Fechter, M. H.; Griengl, H. *J. Mol. Catal. B: Enzym.* **2003**, *24*, 89–98. (e) Huang, S. H.; Tsai, S. W. *J. Mol. Catal. B: Enzym.* **2004**, *28*, 65–69. (f) Sugai, T.; Ohta, H. *Agric. Biol. Chem.* **1991**, *55*, 293–294.
11. (a) Spindler, F.; Pittelkow, U.; Blaser, H. U. *Chirality* **1991**, *3*, 370–375. (b) Chang, C.-Y.; Yang, T.-K. *Tetrahedron: Asymmetry* **2003**, *14*, 2239–2245. (c) Chadha, A.; Manohar, M.; Soundararajan, T.; Lokeswari, T. S. *Tetrahedron: Asymmetry* **1996**, *7*, 1571–1572. (d) Fadnavis, N. W.; Radhika, K. R. *Tetrahedron: Asymmetry* **2004**, *15*, 3443–3447. (e) Schmidt, E.; Blaser, H. U.; Fauquex, P. F.; Sedelmeier, G.; Spindler, F. In *Microbial Reagents in Organic Synthesis*; Servi, S., Ed.; NATO ASI Series C; Kluwer Academic: Dordrecht, 1992; Vol. 381, pp 377–388. (f) Blaser, H. U.; Jalett, H. P. *Stud. Surf. Sci. Catal.* **1993**, *78*, 139–146. (g) Hummel, W.; Schütte, H.; Schmidt, E.; Wandrey, C.; Kula, M. R. *Appl. Microbiol. Biotechnol.* **1987**, *26*, 409–416. (h) Dao, D. H.; Kawai, Y.; Hida, K.; Hornes, S.; Nakamura, K.; Ohno, A.; Okamura, M.; Akasaka, T. *Bull. Chem. Soc. Jpn.* **1998**, *71*, 425–432. (i) Kaluzna, I.; Andrew, A. A.; Bonilla, M.; Martzen, M. R.; Stewart, J. D. *J. Mol. Catal. B: Enzym.* **2002**, *17*, 101–105. (j) Fechter, M. H.; Griengl, H. In Drauz, K., Waldmann, H., Eds. 2nd ed.; Enzyme Catalysis in Organic Synthesis; Wiley-VCH: Weinheim, 2002; Vol. 2, pp 974–989. (k) North, M. *Tetrahedron: Asymmetry* **2003**, *14*, 147–176. (l) Wang, Y.-F.; Chen, S.-T.; Liu, K. K.-C.; Wong, C.-H. *Tetrahedron Lett.* **1989**, *30*, 1917–1920. (m) Herold, P.; Indolese, A. F.; Studer, M.; Jalett, H. P.; Siegrist, U.; Blaser, H. U. *Tetrahedron* **2000**, *56*, 6497–6499.
12. (a) Faber, K. *Chem. Eur. J.* **2001**, *7*, 5004–5010. (b) Strauss, U. T.; Felfer, U.; Faber, K. *Tetrahedron: Asymmetry* **1999**, *10*, 107–117.
13. Huerta, F. F.; Laxmi, Y. R. S.; Bäckvall, J.-E. *Org. Lett.* **2000**, *2*, 1037–1040.
14. Chadha, A.; Baskar, B. *Tetrahedron: Asymmetry* **2002**, *13*, 1461–1464.
15. Although a remarkable number of stereochemically matching pairs of biocatalysts possessing opposite enantio-preference have been reported, the search, for example, 'anti-Kazlauskas' carboxyl ester hydrolases and 'anti-Prelog' alcohol dehydrogenases still represents a major challenge in biocatalysis.

16. Strauss, U. T.; Faber, K. *Tetrahedron: Asymmetry* **1999**, *10*, 4079–4081.
17. For a review on enzyme-catalyzed racemization see: Schnell, B.; Faber, K.; Kroutil, W. *Adv. Synth. Catal.* **2003**, *345*, 653–666.
18. Larissegger-Schnell, B.; Kroutil, W.; Faber, K. *Synlett* **2005**, 1936–1938.
19. For a review on mandelate racemase and its substrate spectrum see: Felfer, U.; Goriup, M.; Koegl, M. F.; Wagner, U.; Larissegger-Schnell, B.; Faber, K.; Kroutil, W. *Adv. Synth. Catal.* **2005**, *347*, 951–961.
20. (a) Glueck, S. M.; Larissegger-Schnell, B.; Csar, K.; Kroutil, W.; Faber, K. *Chem. Commun.* **2005**, 1904–1905. (b) Glueck, S. M.; Pirker, M.; Nestl, B. M.; Ueberbacher, B. T.; Larissegger-Schnell, B.; Csar, K.; Hauer, B.; Stuermer, R.; Kroutil, W.; Faber, K. *J. Org. Chem.* **2005**, *70*, 4028–4032.
21. Microbial degradation of *rac*-**1**,**2** would be expected to occur via (i) enzymatic oxidation catalysed by an NAD(P)H-dependent α-ketoacid reductase/α-hydroxyacid dehydrogenase yielding the corresponding α-ketoacids (phenylpyruvate/4-phenyl-2-oxobutanoate), followed by (ii) enzymatic decarboxylation of the latter catalysed by phenylpyruvate decarboxylase to furnish CO_2 and the corresponding carbaldehydes (phenylacetaldehyde/3-phenylpropionaldehyde. See: Feil, I. K.; Lerch, H.-P.; Schomburg, D. *Eur. J. Biochem.* **1994**, *223*, 857–863. Hummel, W.; Schütte, H.; Kula, M.-R. *Appl. Microbiol. Biotechnol.* **1985**, *21*, 7–15. Vuralhan, Z.; Morais, M. A.; Tai, S.-L.; Piper, M. D. W.; Pronk, J. T. *Appl. Environ. Microbiol.* **2003**, *69*, 4534–4551. Ward, O. P.; Singh, A. *Curr. Opin. Biotechnol.* **2000**, *11*, 520–526.
22. Killian, J. A.; Van Cleve, M. D.; Shayo, Y. F.; Hecht, S. M. *J. Am. Chem. Soc.* **1998**, *120*, 3032–3042.
23. Cohen, S. G.; Weinstein, S. Y. *J. Am. Chem. Soc.* **1964**, *86*, 5326–5330.
24. Chadha, A.; Manohar, M. *Tetrahedron: Asymmetry* **1995**, *6*, 651–652.