Organic & Biomolecular Chemistry

Cite this: *Org. Biomol. Chem.*, 2012, **10**, 3384

www.rsc.org/obc

COMMUNICATION

Synthesis of (*S*)-(+)-cericlamine through lipase-catalyzed aminolysis of azo acetates†

Agnes Prechter,[a] Harald Gröger‡[b] and Markus R. Heinrich*[a]

Received 2nd February 2012, Accepted 8th March 2012
DOI: 10.1039/c2ob25247c

The kinetic enzymatic resolution of azo acetates *via* aminolysis with *Candida antarctica* lipase B has been investigated using benzylamine as amine component. The products obtained from this biotransformation in high enantiomeric purity can serve as valuable precursors for various amino alcohols, as exemplified by the synthesis of the serotonin reuptake inhibitor (*S*)-(+)-cericlamine.

While lipase-catalyzed reactions of tertiary alcohols are usually slow, enzymatic transformations remain fast when primary alcohols with more remote quaternary stereocenters are employed as starting materials.[1] Most primary alcohols that have so far been studied belong to the group of diols **1**,[2] or derivatives in which the unreactive tertiary alcohol is part of an ether,[3] epoxide[4] or lactone.[5] Remarkable results have also been achieved in desymmetrizations of diols **2**.[6] Only very few studies have so far been reported for enzymatic resolutions of primary alcohols **3** bearing a nitrogen-substituted quaternary stereocenter in β-position. An early example is the enantiomeric enrichment of racemic benzamide **4** through lipase AK-20 in the presence of vinyl acetate.[7] A comparable lipase-catalyzed transesterification has very recently been used for the resolution of nitro compound **5** (Fig. 1).[8]

Our interest is focused on azo compounds **6**, which are easily accessible and can also be further converted to manifold products including amino alcohols, amino acids, indolines and isoquinolines.[9,10] The broad substrate scope and substitution pattern of **6** and *O*-acylated derivatives thereof makes an enantioselective approach to such molecules an attractive alternative to currently developed desymmetrization strategies.[6] So far, unfavorably, the access to azo compounds **6** in enantiomerically enriched form has been extremely limited due to the narrow substrate scope, long reaction times and high enzyme loading required for lipase-catalyzed kinetic resolutions.[11] In this communication, we now report recent advances leading to an efficient access to enantiomerically enriched azo acetates through lipase-catalyzed kinetic resolution.

Due to the low conversions observed for azo acetates in the previously employed aqueous biphasic mixtures of methyl *tert*-butyl ether (MTBE) and water,[11] we first examined the hydrolysis of acetate **7a** in a variety of non-dried organic solvents including acetonitrile, MTBE, methanol, *n*-hexane, dimethylformamide (DMF), acetone and tetrahydrofuran (THF) (Table 1, entries 1–8).

The only solvent leading to a basically useful conversion combined with a moderate enantiomeric excess was found to be acetonitrile (entry 1). In a second optimization step benzylamine was added, which turns the overall reaction from a hydrolysis to an aminolysis of the acetate.[12] By this modification, the results for THF, and especially acetonitrile (entries 12, 13), could be further improved with regard to conversion and enantioselectivity. One reason for the positive effect of benzylamine might be the decreased activity of CAL-B otherwise caused by liberated acetic acid.[13] Furthermore, advantages are likely to arise from the ability of benzylamine to act as a better nucleophile than water. Instead of acetic acid, *N*-benzylacetamide is now formed in amounts equivalent to alcohol **8a**.[14] While fewer equivalents of benzylamine (3 equiv) did not influence the overall outcome (entry 14), larger quantities of benzylamine (>5 equiv) as well as the addition of water led to remarkably lower conversions

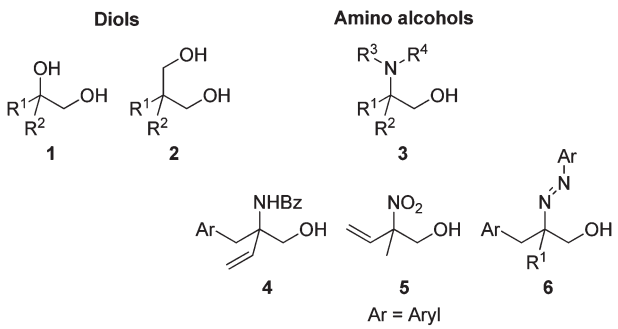


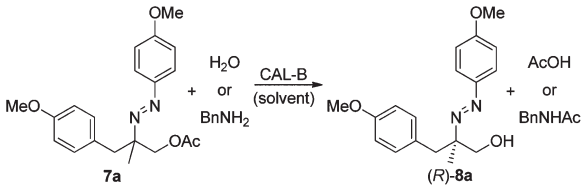
Fig. 1 Substrates for kinetic enzymatic resolution.

[a]*Department für Chemie und Pharmazie, Pharmazeutische Chemie, Friedrich-Alexander Universität Erlangen-Nürnberg, Schuhstraße 19, 91052 Erlangen, Germany. E-mail: Markus.Heinrich@medchem.uni-erlangen.de; Fax: +49-9131-85-22585; Tel: +49-9131-85-24115*
[b]*Department für Chemie und Pharmazie, Organische Chemie, Friedrich-Alexander-Universität Erlangen-Nürnberg, Henkestraße 42, 91054 Erlangen, Germany*

†Electronic supplementary information (ESI) available: Experimental procedures, characterization data and copies of ^{1}H and ^{13}C NMR spectra of all new compounds. See DOI: 10.1039/c2ob25247c

‡Current address: Organische Chemie I, Fakultät für Chemie, Universität Bielefeld, 33501 Bielefeld, Germany. E-mail: harald.groeger@uni-bielefeld.de.

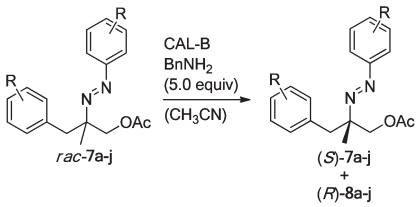
Table 1 Effects of non-polar solvents and benzylamine



Entry[a]	Solvent	Base (equiv)	Conversion[b]	% ee (**8a**)[b]	E[c]
1	CH_3CN	—	32	41	2.9
2	CH_3CN[d]	—	<2	nd[e]	nd[e]
3	MTBE	—	67	4[f]	1.1
4	MeOH	—	<1	nd[e]	nd[e]
5	*n*-Hexane	—	21	0	1.0
6	DMF	—	<1	nd[e]	nd[e]
7	Acetone	—	13	45	2.8
8	THF	—	13	35	2.2
9	MTBE	$BnNH_2$ (5)	45	18	1.6
10	DMF	$BnNH_2$ (5)	<1	nd[e]	nd[e]
11	Acetone	$BnNH_2$ (5)	16	42	2.6
12	THF	$BnNH_2$ (5)	28	43	2.9
13	**CH_3CN**	**$BnNH_2$ (5)**	**45**	**62**	**7.0**
14	CH_3CN	$BnNH_2$ (3)	46	62	7.1
15	CH_3CN	$BnNH_2$ (7)	35	64	6.3
16	CH_3CN	$BnNH_2$ (10)	21	51	3.5
17	CH_3CN[g]	$BnNH_2$ (5)	<2	nd[e]	nd[e]
18	CH_3CN	*n*-$BuNH_2$ (5)	31	76	10.2
19	CH_3CN	Et_3N (5)	32	46	3.3
20	CH_3CN	Et_2NH (5)	19	68	6.1

[a] Reaction conditions: *rac*-**7a** (0.05 mmol), organic solvent (2 mL), CAL-B (20 mg), rt, 3 days. [b] Conversion and enantiomeric excess of **8a** determined by chiral HPLC. [c] E-value calculated from the conversion and the ee data of the alcohol **8a**. [d] Water was added to the reaction mixture: CH_3CN–H_2O 3 : 1(v/v). [e] nd = not determined. [f] (*S*)-enantiomer. [g] 5% water were added to the reaction mixture.

Table 2 Variation of substrates



Entry[a]	R	Acetate (*S*)-**7**[c,d] (%, ee)	Alcohol (*R*)-**8**[c] (ee)	E[e]
1[b]	4-OMe	**7a** (84, 73% ee)[f]	**8a** (53% ee)	6.8
2	4-F	**7b** (58, 93% ee)	**8b** (40% ee)	7.1
3	3,4-Cl_2	**7c** (52, >99% ee)	**8c** (37% ee)	9.6[g]
4	4-Br	**7d** (58, 96% ee)	**8d** (35% ee)	7.0[g]
5	H	**7e** (24, 84% ee)	**8e** (13% ee)	2.7
6	2-OMe	**7f** (n.d.)[h]	**8f** (18% ee)[h]	1.5[i]
7	3-OMe	**7g** (82, 56% ee)	**8g** (32% ee)	3.2
8	3,4-OMe_2	**7h** (n.d.)[h]	**8h** (71% ee)[h]	7.3[i]
9	4-CN	**7i** (74, 83% ee)	**8i** (43% ee)	6.0
10[j]	4-NO_2	**7j** (48, 90% ee)	**8j** (29% ee)	4.8

[a] Reaction conditions: *rac*-**7** (0.30 mmol), CH_3CN (12 mL), CAL-B (120 mg), benzylamine (161 mg, 5 equiv), rt, 3 days. [b] This reaction was performed on a 0.05 mmol scale for 6 days. [c] Enantiomeric excess determined by chiral HPLC, yields are calculated based on the single enantiomers. [d] Acetates were converted to their corresponding alcohols for HPLC-measurement except in the case of **7j**, which could be measured directly. [e] E-value calculated from the ee data of the alcohol and the acetate. [f] Enantiomeric excess calculated from conversion and enantiomeric excess of the alcohol. [g] E-values are lower than those reported in Table 3 due to continuation of the reaction beyond the optimal conversion. [h] Enantiomeric excess of **7f** and **7h** not determined due to low conversions (30% for **7f** and 24% for **7h**, each after 3 days). [i] E-values calculated from the conversion and the ee data of the alcohol. [j] This reaction was performed on a 0.06 mmol scale.

(entries 15–17). Attempts with other bases instead of benzylamine revealed that *n*-butylamine as an additive is able to give even higher E-values, but at remarkably lower conversions (entry 18). The reaction with triethylamine proceeded as if no base had been added (entry 19, *cf.* entry 1), and diethylamine led to an improved ee-value, but with insufficient conversion (entry 20).

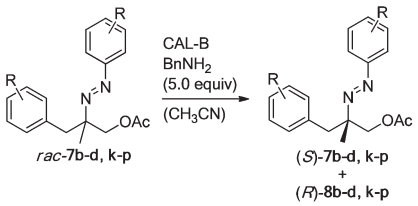
The newly found conditions (Table 1, entry 13) were then applied to the fluorinated azo acetate **7b**, a compound which had shown the worst results in previous investigations.[11] To get a better insight in the conversion of each enantiomer, the reaction course was followed by chiral HPLC over several days (see ESI†).[15,16] In this way, nearly total conversion of one enantiomer was reached after 72 hours, leaving the remaining acetate (29% of 50% theoretical yield) with necessarily high enantiomeric excess.

Results from reactions with a broader spectrum of substrates are summarized in Table 2. After 3 days, high ee-values were observed for the remaining acetates **7** starting from racemic lipophilic halogenated azo acetates **7b–d** (entries 2–4). While a longer reaction time of 6 days was necessary to achieve an acceptable enantiomeric excess for **7a** (entry 1), the conversion of unsubstituted azo acetate **7e** proceeded rapidly, but with only low selectivity (entry 5). In general, electron-donating substituents such as the methoxy group led to the lowest conversions when situated in 2- or 4-position of the aromatic core (entries 1, 6, 8). A higher conversion, although at lower selectivity, was reached with the 3-methoxy derivative **7g** (entry 7). Electron-withdrawing substituents like nitro or cyano were better tolerated in the 4-position than the methoxy group, as shown by the reactions of **7i** and **7j** (entries 9, 10). The fact that the desired products (*S*)-**7i** and (*S*)-**7j** were still obtained with lower enantiomeric excess than the halogenated derivatives demonstrated that the latter group of substrates (entries 2–4) represents the most suitable for this type of enzymatic resolution.[17]

The results obtained from the investigation of the substrate scope (Table 2) raised the question whether high ee-values (>95%) can especially be reached with halogenated azo acetates at relatively low conversions. A survey within this group of compounds is presented in Table 3. Interestingly, the conversion of some azo acetates **7** required much shorter reaction times than three days to yield the desired products with acceptably high enantiomeric excess.

When gradually increasing the steric demand of the halogen atom in the 4-position from fluorine to chlorine and bromine to iodine, a constant decrease in reaction time was observed (entries 1–7). To reach roughly 90% ee with a nearly similar yield, the 4-fluoro derivative **7b** required 48 hours, the 4-chloro derivative **7k** 44 hours, the 4-bromo derivative **7d** 33.5 hours and 4-iodo derivative **7l** only 24.5 hours (entries 1, 3, 5, 6).

Table 3 Variation of substrates and reaction times



Entry[a]	R	Time (h)	Acetate (*S*)-**7**[b,c] (%, ee)	Alcohol (*R*)-**8**[b] (ee)	*E*[d]
1	4-F	48	**7b** (70, 88% ee)[e]	**8b** (48% ee)	7.7
2	4-F	71	**7b** (58, 96% ee)[e]	**8b** (39% ee)	7.8
3	4-Cl	44	**7k** (70, 90% ee)	**8k** (50% ee)	8.6
4	4-Cl	59	**7k** (64, 94% ee)	**8k** (43% ee)	8.0
5	4-Br	33.5	**7d** (72, 91% ee)	**8d** (51% ee)	9.1
6	4-I	24.5	**7l** (76, 90% ee)	**8l** (52% ee)	9.1
7	4-I	46.5	**7l** (66, >97% ee)	**8l** (48% ee)	10.8
8	2-Br	149	**7m** (66, 60% ee)	**8m** (32% ee)	3.4
9	3-Br	16	**7n** (60, 82% ee)	**8n** (37% ee)	5.0
10	3-Br	45	**7n** (22, 96% ee)	**8n** (12% ee)	3.5
11	2,4-Cl_2	39	**7o** (66, 80% ee)[f]	**8o** (nd)[g]	5.2[h]
12	3,5-Cl_2	12.5	**7p** (74, 89% ee)[i]	**8p** (51% ee)[i]	8.6
13	3,4-Cl_2	15	**7c** (80, 97% ee)	**8c** (64% ee)	18.1
14	3,4-Cl_2	19	**7c** (74, >99% ee)	**8c** (59% ee)	18.8

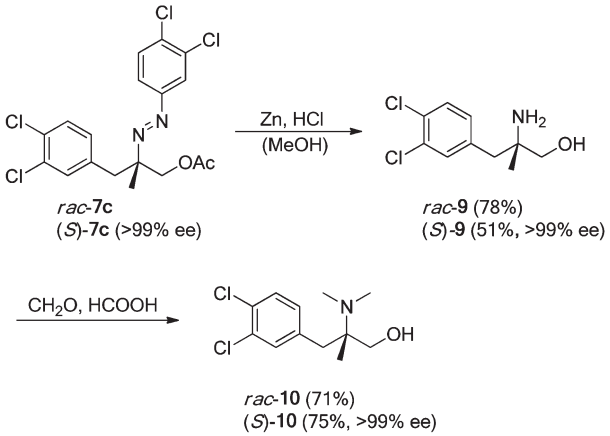
[a] Reaction conditions: *rac*-**7** (0.20 mmol), CH_3CN (8 mL), CAL-B (80 mg), benzylamine (107 mg, 5 equiv), rt. [b] Enantiomeric excess determined by chiral HPLC, yields are calculated based on the single enantiomers. [c] Unless otherwise stated, acetates were converted to their corresponding alcohols for HPLC-measurement. [d] *E*-value calculated from the ee data of the alcohol and the acetate. [e] Enantiomeric excess calculated from conversion and enantiomeric excess of the alcohol. [f] Determination of enantiomeric excess after conversion to phthalate derivative. [g] nd = not determined. [h] *E*-value calculated from the conversion and the ee data of the alcohol. [i] Determination of enantiomeric excess after conversion to amino alcohol.

Shifting the rather large bromo substituent into the 2- or 3-position led to a sharp decrease (entry 8) or increase (entries 9, 10) in reaction rate, respectively. Both compounds **7m** and **7n** were however converted with comparably lowered *E*-values. The substituent effects observed for the three different dichloro derivatives **7c**, **7o** and **7p** are largely in agreement with the observations made before. While a 2-substituent such as in **7o** (entry 11) is not well tolerated even at small size, 3-substitution as in **7p** (entry 12) allows for better results. As evidenced by the experiments with **7c** (entries 13, 14), two small and lipophilic substituents in 3- and 4-position are likely to be one of the most suitable patterns for the enzymatic reaction described herein.[18]

One possible application for azo acetates obtained from the kinetic resolution is the synthesis of pharmaceutically relevant amino alcohols. (*S*)-(+)-Cericlamine [(*S*)-**10**], an antidepressant,[19,20] is readily accessible from acetate (*S*)-**7c** through two simple synthetic steps (Scheme 1).

Reduction of azo acetate **7c** to amino alcohol **9** by zinc in hydrochloric acid was followed by reductive methylation.[21] In addition to the experiments reported in Table 3 (entries 13, 14), the enzymatic resolution of **7c** was carried out on a larger scale to provide 0.84 g (76% yield based on single enantiomer, 97% ee) of azo compound (*S*)-**7c**.

In summary, azo acetates have been shown to be valuable starting materials for the enantioselective preparation of diverse



Scheme 1 Synthesis of (*S*)-(+)-cericlamine [(*S*)-**10**].

(*S*)-2-amino-3-phenylpropanols. The results obtained by enzymatic kinetic resolution compare well to existing synthetic strategies leading to structurally related amino alcohols.[7,8] The tolerance of lipophilic substrates enabled an effective synthesis of (*S*)-(+)-cericlamine, and *E*-values up to 18 were observed in the enzymatic transformation leading to the required (*S*)-(+)-cericlamine intermediate. Ongoing research is directed towards dynamic kinetic resolutions[22] as well as further process optimization.

Acknowledgements

The authors would like to thank the Universität Bayern e.V. for a "Bayerische Eliteförderung" fellowship awarded to A. Prechter. We thank Michael Höfer for the experiments conducted during his pharmaceutical chemistry internship.

Notes and references

1 (*a*) U. T. Bornscheuer and R. J. Kazlauskas, *Hydrolases in Organic Synthesis*, Wiley-VCH, 2006, 2nd edn; (*b*) For a recent review discussing lipase-catalyzed reactions see: A. Ghanem, *Tetrahedron*, 2007, **63**, 1721.
2 (*a*) S. T. Chen and J. M. Fang, *J. Org. Chem.*, 1997, **62**, 4349; (*b*) R. P. Hof and R. M. Kellog, *J. Chem. Soc., Perkin Trans. 1*, 1996, 2051; (*c*) R. P. Hof and R. M. Kellog, *Tetrahedron: Asymmetry*, 1994, **5**, 565; (*d*) C. R. Johnson, Y. Xu, K. C. Nicolaou, Z. Yang, R. K. Guy, J. G. Dong and N. Berova, *Tetrahedron Lett.*, 1995, **36**, 3291; (*e*) V. P. Rocco, S. J. Danishefsky and G. K. Schulte, *Tetrahedron Lett.*, 1991, **32**, 6671–6674; (*f*) O. Jimenéz, M. P. Bosch and A. Guerrero, *J. Org. Chem.*, 1997, **62**, 3496; (*g*) V. Khlebnikow, K. Mori, K. Terashima, Y. Tanaka and M. Sato, *Chem. Pharm. Bull.*, 1995, **43**, 1659; (*h*) K. E. Henegar, S. W. Ashford, T. A. Baughman, J. C. Sih and R. L. Gu, *J. Org. Chem.*, 1997, **62**, 6588; (*i*) J. C. Sih, *J. Am. Oil Chem. Soc.*, 1996, **73**, 1377; (*j*) R. G. Lovey, A. K. Saksena and V. M. Girijavallabhan, *Tetrahedron Lett.*, 1994, **35**, 6047.
3 (*a*) H. Estermann, K. Prasad, M. J. Shapiro, J. J. Bolsterli and M. D. Walkinshaw, *Tetrahedron Lett.*, 1990, **31**, 445; (*b*) B. Wirz, R. Barner and J. Huebscher, *J. Org. Chem.*, 1993, **58**, 3980; (*c*) R. P. Hof and R. M. Kellog, *J. Org. Chem.*, 1996, **61**, 3423; (*d*) J. A. Hyatt and C. Skelton, *Tetrahedron: Asymmetry*, 1997, **8**, 523.
4 (*a*) P. Ferraboschi, D. Brembilla, P. Grisenti and E. Santaniello, *J. Org. Chem.*, 1991, **56**, 5478; (*b*) P. Ferraboschi, S. Casati, P. Grisenti and E. Santaniello, *Tetrahedron: Asymmetry*, 1993, **4**, 9; (*c*) P. Ferraboschi, S. Casati, P. Grisenti and E. Santaniello, *Tetrahedron*, 1994, **50**, 3251; (*d*) P. Ferraboschi, P. Grisenti, S. Casati and E. Santaniello, *Synlett*, 1994, 754; (*e*) P. Ferraboschi, P. Grisenti, A. Manzocchi and E. Santaniello, *Tetrahedron: Asymmetry*, 1994, **5**, 691; (*f*) Y. B. Seu and Y. H. Kho, *Tetrahedron Lett.*, 1992, **33**, 7015; (*g*) T. Itoh, H. Ohara, Y. Takagi,

N. Kanada and K. Uneyama, *Tetrahedron Lett.*, 1993, **34**, 4215; (*h*) See ref. 2*a*.
5 T. Tauchi, H. Sakuma, T. Ohno, N. Mase, H. Yoda and K. Takabe, *Tetrahedron: Asymmetry*, 2006, **17**, 2195.
6 (*a*) M. Ihara, M. Suzuki, K. Fukumoto and C. Kabuto, *J. Am. Chem. Soc.*, 1990, **112**, 1164; (*b*) N. Watanabe, T. Sugai and H. Ohta, *Chem. Lett.*, 1992, 657; (*c*) S. Akai, T. Naka, Y. Takebe and Y. Kita, *Tetrahedron Lett.*, 1997, **38**, 4243; (*d*) A. Fadel and P. Arzel, *Tetrahedron: Asymmetry*, 1997, **8**, 283 and 371 (*e*) T. Ping, M.-H. Xu, Z.-Q. Wang, Z.-Y. Li and G.-Q. Lin, *Synlett*, 2006, 1201; (*f*) S. Sano, M. Nakao, M. Takeyasu, T. Honjo and Y. Nagao, *Lett. Org. Chem.*, 2006, **3**, 764.
7 D. B. Berkowitz, J. A. Pumphrey and Q. Shen, *Tetrahedron Lett.*, 1994, **35**, 8743.
8 T. Nakano, Y. Yagi, M. Miyahara, A. Kamimura, M. Kawatsura and T. Itoh, *Molecules*, 2011, **16**, 6747.
9 For related review articles, see: (*a*) M. R. Heinrich, *Chem.–Eur. J.*, 2009, **15**, 820; (*b*) S. B. Höfling and M. R. Heinrich, *Synthesis*, 2011, 173.
10 (*a*) M. R. Heinrich, O. Blank and S. Wölfel, *Org. Lett.*, 2006, **8**, 3323; (*b*) M. R. Heinrich, O. Blank and A. Wetzel, *J. Org. Chem.*, 2007, **72**, 476; (*c*) O. Blank, A. Wetzel, D. Ullrich and M. R. Heinrich, *Eur. J. Org. Chem.*, 2008, 3179.
11 F. R. Dietz, A. Prechter, H. Gröger and M. R. Heinrich, *Tetrahedron Lett.*, 2011, **52**, 655.
12 For some enzymatic aminolysis reactions, see: (*a*) S. Puertas, R. Brieva, F. Rebolledo and V. Gotor, *Tetrahedron*, 1993, **49**, 4007; (*b*) A. E. Sigmund, K. C. McNulty, D. Nguyen, C. E. Silverman, P. Ma, J. A. Pesti and R. DiCosimo, *Can. J. Chem.*, 2002, **80**, 608; (*c*) M. López-Garcia, I. Alfonso and V. Gotor, *J. Org. Chem.*, 2003, **68**, 648; (*e*) M. Weiß and H. Gröger, *Synlett*, 2009, 1251; (*e*) M. Weiß, T. Brinkmann and H. Gröger, *Green Chem.*, 2010, **12**, 1580; (*f*) Review:. V. Gotor, *J. Biotechnol.*, 2002, **96**, 35.
13 M. D. Romero, L. Calvo, C. Alba and A. Daneshfar, *J. Biotechnol.*, 2007, **127**, 269.
14 For details, see ESI.†
15 Conversions determined by HPLC were verified by ^{1}H-NMR spectra. According to ^{1}H-NMR, all reactions remained free of side-products.
16 Reaction conditions: *rac*-**7b** (0.20 mmol), CH_3CN (8 mL), CAL-B (80 mg), benzylamine (107 mg, 5 equiv), rt.
17 *R*/*S*-selectivity in agreement with previous studies, see ref. 11.
18 Experiments performed with two different enzyme batches led to nearly identical results.
19 (*a*) G. Aubard, J. Bure, A. P. Calvet, C. Gouret, A. G. Grouhel and J.-L. Junien, *Eur. Pat*, 0237366, 1987; (*b*) J. G. Wettstein, C. J. Gouret and J. L. Junien, *Eur. J. Pharmacol.*, 1990, **183**, 1477; (*c*) C. J. Gouret, J. G. Wettstein, R. D. Porsolt, A. Puech and J.-L. Junien, *Eur. J. Pharmacol.*, 1990, **183**, 1478; (*d*) C. J. Gouret, R. Porsolt, J. G. Wettstein, A. Puech, X. Pascaud and J.-L. Junien, *Arzneim.-Forsch.*, 1990, **40**, 633.
20 For other enantioselective synthetic approaches to cericlamine, see: (*a*) B. Kaptein, H. M. Moody, Q. B. Broxterman and J. Kamphuis, *J. Chem. Soc., Perkin Trans. 1*, 1994, 1495; (*b*) A. G. Steinig and D. M. Spero, *J. Org. Chem.*, 1999, **64**, 2406.
21 The configuration of the product was confirmed by comparison of the optical rotation with reported values, see ref. 20*a*.
22 For possible light-induced racemization of quaternary azo compounds see:N. A. Porter and L. J. Marnett, *J. Am. Chem. Soc.*, 1973, **95**, 4361.